

Training Technicians for the Future

THE British government seems on the point of dismantling the apparatus developed in the past few years for industrial training, and although there are several reasons why people should have their knives into the industrial training boards the open question now is whether the British government will take seriously the problems of industrial training and the need to provide a sufficient incentive for the continuing improvement of skill at all levels of industry and commerce. This week, the Engineering Industry Training Board has published its reply to the government's proposals set out some months ago under the heading, "Training for the Future", and, because the board argues for a continuation of the present arrangements under which levies are collected from companies on the basis of turnover and then dispensed as grants to those who provide training for their employees, it is bound to be regarded as a piece of special pleading. Before the whole machinery of the industrial training boards is dismantled, however, the British government and those who pay its taxes should give some thought to the arrangements that will be needed in the years ahead if industrial training is to be efficient.

In retrospect, it is clear that the most serious defect of the industrial training boards has been their concern with throughput rather than with quality. The Engineering Industry Training Board has been, from the beginning (in 1964), one of the largest. For several years, it has been collecting more than £75 million a year in levies, amounting to between 2.2 and 2.5 per cent of the total payroll of the engineering industry, and has been distributing all but a few million pounds of this as grants to those companies which chose to send people on courses to improve their skills or which undertook to provide training of some kind or another on the premises. The board itself, like many others in the field, has also dabbled with the development of training programmes. It is in particular responsible for the development of a gargantuan but mediocre series of training manuals which are as extravagant in their presentation as they are flatulent in content and pedagogically offensive. Few educationalists would regard this venture as permanently useful. Most users have come, with the passage of time, to think of it as the kind of penance which inevitably accompanies a grant-giving system which is too arithmetical for intellectual comfort. But it is only fair to say that other industrial training boards have been even more extravagant than the Engineering Industry Training Board in their expectations of how much can be done by interlocking committees of industrialists and trade unionists to improve the methods of industrial training. The Catering Industry Training Board has an exceedingly naive collection of curricula to its discredit. But there have also been industrial training boards—the Construction Industry Training Board, for example—which have so mismanaged their affairs that the process of robbing Peter to pay Paul has got embarrassingly out of hand.

It would have been more prudent for the training

boards to have set about their task in a more modest fashion. The truth is that industrial training is intellectually as difficult a task as any other kind of training. In 1964, when the Labour government passed the Industrial Training Act, it was right and proper that people should wring their hands over the neglect of training. It was entirely wrong, however, of the Labour government to suppose that sheer parsimony was the cause of trouble. To be sure, some industrial companies, neglectful of their own long-term interests as well as those of their employees, made too little use of the opportunities for the improvement of the skill of craftsmen and technicians that were then available. Even in such well-known fields as machine tool operation, however, the means available in 1964 for the assessment of the value of particular courses of training were rudimentary, to say the best of them. Would a craftsman benefit more from a little potted physics and mechanical engineering than from painstaking instruction on the job? Might not much depend on the different aptitudes of different people? These questions have never been seriously asked, no doubt because of the difficulties that would arise from any suggestion that even on the shop floor, some people are regrettably more equal than others. And outside the fields of training which have become pedagogically well developed, the industrial training boards have chosen to operate in a kind of shifting sand. The proliferation in the past few years of second-rate management courses has no doubt been a great boon to those employees who have been impelled by either ambition or the wish for a little extra leisure to spend weeks at a time or even longer in elegant country houses, but it would require a more intricate economic analysis than any so far carried out to tell whether the result has been an improvement of industrial efficiency or the opposite.

The Engineering Industry Training Board's response to the government's proposals is an unsatisfactory half-way house. Apparently recognizing that it will be hard entirely to head the government away from its intention of replacing the system of grants and levies by one in which finance is provided from central funds, the board advocates a middle course, with something like 0.5 per cent of the engineering industry's payroll collected by means of levies and an extra £20 million a year provided by the government. The essence of the case for this proposal is the board's contention that financial incentives to individual companies are necessary to persuade at least the black sheep among them that industrial training is an important and even an urgent need. The board goes some way to acknowledge the force of one of the objections which have been made to its present method of operation, that companies with well-developed schemes for training can hope to earn more by training grants than they pay in levies, by promising that if it is allowed to continue to exist, it would "disengage" from those companies which had already become efficient. But who is to say whether smaller companies might not seriously



calculate that it is better to live by the skills which they are able to recruit, possibly after a deliberate decision to spend more on salaries, than to spend time and money on training? The board, in its statement of its own proposals, complains that the engineering industry's interest in training has by no means been consolidated, and cites as evidence the way in which, last year, when employment in the engineering industry declined by five per cent, the amount of energy spent on training declined by 20 per cent. But may it not be that these economies by industrial firms were a measure of their discontent with industrial training as at present organized rather than a measure of fecklessness?

The central feature of the British government's proposals on industrial training are that there should be a National Training Agency and, at this stage, it remains to be seen precisely what relationship there will eventually be between the agency and the boards. The strongest argument for a central direction of affairs is, however, that industrial training, like other forms of education, should be a national responsibility. Once governments are committed to the view that industrial and commercial organizations should be responsible for the cost of training their own employees, they are quickly caught up in questions of whether employers can or should be expected to equip employees for better jobs elsewhere and whether, in any case, training conducted on a company basis is likely to provide the kind of flexibility that will in due course suit the national interest. If, one way or another, the government can devise a way of channeling substantial support for industrial training through the Industrial Training Agency, it will have gained a substantial benefit. If it has sense, the government will seek to use the vast apparatus of institutions for further education as a vehicle for training. This has for too long been a neglected sector of British education. The biggest danger, in the months ahead, is that the government will fail to incorporate in its new proposals a sufficiently radical approach to the educational research and curriculum development on which industrial training might be more soundly founded. But there are also dangers that the government will spend too little and that it will concentrate on types of training which are comparatively well understood. For all their faults, the industrial training boards did at least have an incentive to tackle new kinds of training which were, by definition, more difficult.

New Statesmanship

NOBODY should take pleasure in the events which have, in the past few months, attended the sad metamorphosis of the *New Statesman*. For donkey's years that other weekly journal has been a powerful focus of radical expression. On the face of things, Mr Richard Crossman would have been a splendid inheritor of Mr Kingsley Martin's legacy, a journal of controlled subversion. In the event—and nobody should complain—he was too old, and by a long chalk far too busy. The result was that the *New Statesman* became what it always might have become, a journal of prejudice, not of opinion. There is a limit to the extent to which it is permissible for editors to write as if the centre of the world is where they are, and in particular there is a limit to the patience

of the world at large with the doings of and the goings-on within the British Parliamentary Labour Party. So much, by unfortunate demonstration, is now self-evident. It will no doubt be a perpetual cause for discontent that Mr Crossman seems not to have found it possible to import into his conduct of the *New Statesman*'s affairs the urbanity which has made him one of the best of all literary critics, especially on German affairs.

What will happen now? On the face of things, the outlook is not cheerful. Mr Anthony Howard, Mr Crossman's successor, is a political animal. He is the kind of fellow whose strength is that he can quickly tell which way the wind is blowing. To acknowledge, as Mr Howard has apparently done, that further opposition to the European venture on which Britain is now engaged is useless may be sensible enough, for that is how the dice are stacked, but even those who applaud what may seem in some quarters to be a death-bed conversion will await with interest and with some alarm the development of this unfashionable but laudable line of argument. Will Mr Howard and his colleagues address themselves to a radical examination of what the European Community should be like, or will they continue to reflect, as Mr Crossman has done, conventional prejudices about what the Left requires of the future? Only time, alas, will tell. Comparatively passive (for the time being) weekly journals such as *Nature* must of necessity be sparing with advice. It remains, however, a simple fact that the most obvious gap in contemporary radical journalism is reporting. Telling it like it is is at once too difficult and too expensive. The result is that most weekly journals in Britain have fallen back on opinion, *sui generis*, which has at least the advantage of being cheaper. So is it any wonder that the *New Statesman*, like many other journals with the same constituency, has fallen back on simple declarations of belief? Mr Howard, by all accounts, is his colleagues' man. His first task will be to persuade them that accounts of the world as it is are as powerful an argument for change as any theoretical doctrine.

100 Years Ago



NOTES

AFTER we went to press last week, a most cheering telegram was received in this country respecting the fate of Dr. Livingstone, as follows:—'Aden, May 1, 1872.—The *Abydos* has returned from Zanzibar. She brings news that Dr. Livingstone is safe with Stanley. The news is brought by natives.' It may fairly be hoped that still more authentic intelligence will shortly be received respecting the fate of the great traveller, with respect to whom such anxiety has been manifested in this country. Another despatch speaks also of the destruction of a large portion of the town of Zanzibar by a terrible hurricane on April 15.

From *Nature*, 6, 32, May 9, 1872.

OLD WORLD

Brighter Future for the Royal Institution

AFTER restricting the deficit of the Royal Institution to less than £10,000 a year during each of the past three years, inflation has finally got the best of all efforts and a deficit of £17,600 is forecast for 1972.

In drastic efforts to solve the institution's financial problems the managers have taken the unprecedented step of introducing a charge on all members from January 1973 for attendance at the evening discourse meetings. Attendance at these discourses has until now been free to members—although guests have had to pay a charge since 1969—but subject to approval at a special general meeting to be held in July it will cost members 60p on each occasion they attend the main theatre—or 30p if they prefer to sit in the gallery. Members will, however, be able to buy a season ticket for £6.65 which will enable them to attend all of the twenty discourses held in the year.

By instituting such charges the managers have avoided the potentially dangerous effects of increasing subscriptions which could have resulted in a drop in membership, thus reversing the trend of 1971 which saw a net increase of 243, with 314 new members having enrolled during the year bringing the total membership to 1,483.

The Royal Institution expects, if the average attendance at discourses remains the same, that the new moves will raise about £4,000 a year. This, however, will not by itself solve the financial problems of the institution. Other efforts to improve the situation include a campaign to make the library self sufficient by selling books which are judged not to be relevant to the purposes of the institution. So far this has raised about £100,000 and Sir Gordon Cox, the treasurer of the Royal Institution, said this week that the target is £150,000 which, if invested, could provide an annual income of £7,500.

In the institution's annual report published recently Sir Gordon Cox attributes the smaller deficit for 1971 (£7,812 compared to £9,017 in 1970) chiefly to economies carried out by the staff. These economies were, however, mostly wiped out by salary increases of £10,000 which were necessary according to the report to "ensure that the remuneration of the staff is kept in line with that for comparable employment elsewhere in London".

Another item of expenditure which appears on this year's balance sheet but

did not last year is a sum of £2,001 for rates. In the past the rates have been paid through a government grant but the government will not in future pay for annual increases in rates although it will still reimburse the institution for the bulk of the payment to the local authority.

The treasurer warns that continued economies will be necessary and that every possible avenue must be explored in the search for more income. The property owned by the institution needs maintenance and Sir Gordon points out that even if this is phased over several years it is still a very formidable proposition.

In a plea for more sponsorship of its activities Sir Gordon points out that the gap between income and expenditure on the institution's educational activities was closed to some extent last year by the Courtauld Educational Trust which

agreed to sponsor a series of school lectures for five years. He added that he hoped that this example will "be followed by others so that this important side of our work may become self-sufficient financially".

Certainly with Sir Gordon Cox at the helm the future for the Royal Institution, although it may not be too rosy for the coming year, does look a lot healthier.

Sir Gordon Cox said this week that he was hopeful that by the beginning of 1973 the financial position of the Royal Institution will be improved but that 1972 will "strain the resources". As for the future, Sir Gordon said that he was very hopeful that in a few years the balance sheet will show a surplus but that this will depend on maintaining the momentum of the increased membership drive that has showed such dividends in the past 16 months.

SCIENCE RESEARCH COUNCIL

More Space at RSRS

THE Science Research Council's Radio and Space Research Station at Slough is to expand its space activities, according to the report of the Radio and Space Research Station review panel published last week (Science Research Council, 1972). One of the chief changes, which was implemented in February this year, is the removal of the Space Research Management Unit from the council's London office to the RSRS, thus centralizing much of the SRC's support for space science at Slough.

Before February, the budget of the RSRS was about £1.3 million a year, but the management unit has brought with it an extra £3 million a year, most of which is for the purchase of Skylark, Petrel and Skua rockets, satellites and services for the SRC's space programme. The panel does, however, recommend that some of the basic radio research activities should be cut back during the next two to three years to make way for the station's increasing space commitment. Although the panel thinks it important that research on tropospheric physics and propagation at centimetre and millimetre wavelengths "should be continued at about its present level", it recommends that the ionospheric physics programme should be halved during the next two or three years and should in future concentrate on the improvement of ionospheric communi-

cations and the continuing provision of an advisory service. The number of staff involved in basic radio research is expected to drop from 86 in 1970/71 to 60 in 1973/74, and the funding to be reduced from £550,000 to £425,000 during the same period.

Dr J. R. Saxton, director of the RSRS, emphasized this week that the station was in no sense turning into an establishment devoted solely to the provision of services, and that basic research would continue to consume about 30 per cent of the station's effort. The panel also suggested that research on a contract basis might sometimes be undertaken for outside organizations, but Dr Saxton said that such contract work would have to be carefully selected because of the problems of providing the necessary staff, especially for short-term contracts.

The report also has something to say about the relationship between the RSRS and space and radio groups in universities. It says, for example, that "thought is being given" to the idea of using the station as a centre for back-up services for university space groups, especially the development of instrumentation at present being done by establishments like the Rutherford Laboratory. The panel also considers that the station should encourage universities to collaborate in research on topics such as the influence of meteorological factors on microwave propagation to which they seem at present to pay scant attention.

RESEARCH COUNCILS

Letting the World Know

DURING the tortuous debate on Lord Rothschild's green paper *A Framework for Government Research and Development* the research councils were at times accused not so much of not doing good work, as not being seen to be doing it. If Lord Rothschild had started work later, or the Department of Education and Science earlier that accusation might not have been made, for the DES has just produced like a rabbit from a hat seven films and £70,000 worth of exhibition about the work of the research councils.

The exhibition, entitled *Search*, which opened on Monday at the Science Museum is the fruit of three years work and will run until October. Designed by Mr James Gardner—the man with QE2 and the British pavilion at Expo '67 to his name—the exhibition is a series of chambers that lead deeper and deeper into the realms of science. The exhibition starts in darkest night with the world of astronomy. The sky at night looms up as seen by the naked eye and then by a radio telescope, with pulsars pulsing and old stars dying. A bank of electronic gadgetry illustrates the principles of aerial synthesis, the possible formation of the universe and the use of colour in astronomy. Overhead, Aerial 4 chatters away above a list of the experiments on board while on the far side of the room a model of the Rutherford laboratory's Nimrod accelerator whizzes protons round to send them crashing into a model bubble chamber while slides overhead explain stage by stage the interactions that take place.

The, by now, fascinated spectator is led to the section on science and the environment with the Earth and marine sciences on display. Relief globes, antarctic landscapes and moving plates rub shoulders, while GLORIA the National Institute of Oceanography's mapping device blips away tracing the sea bed. Across the gangway leafless peas and strawberries that grow strawberry-side up show how plants are being adapted to harvesting machines, and jars of plankton demonstrate why the herring left Plymouth in the 1930s only to start to return last year. Samples of soil and geological mapping come next as visitors walk under the outstretched roots of an apple tree.

A model cow with bellows for lungs and bagpipes and Cona coffee jugs for stomachs wheezes away, while other exhibits demonstrate multiovalulation in sheep and synthetic control of insects. The surroundings change to a vivid pink and a gigantic rubber heart pumps disturbingly away in its glass cage, while on a telephone a voice says "there

follows a rapid irregular heart beat" as an electrocardiogram registers frenetic leaps of activity.

The normal functioning of cells is illustrated by the action of nerve, heart and muscle cells and an enormous spermatozoon precedes a display of genetic and chromosome abnormalities. Work on cancer research is illustrated by a series of slides and linked sound commentary. There is much more besides with exhibits on the hydrological cycle, artificial limbs and nitrogen fixation for beginners. There is an immense sense of bustling activity kept alive by a refreshing irreverence. Cartoons are sprinkled about the place and a dressed up dolly bird quivers in a gilded cage beneath the sign "female in full mating plumage".

Studiedly non-technical, *Search* provides much information for those prepared to read their way right round. For those who are not, it provides a formidable display of some of the activities of those mysterious bodies the research councils. For the Science Museum, it's the best exhibition they've opened recently and for the research councils it's a good piece of public relations: science is relevant to man's needs as well as being fun.

The same cannot be said with much conviction for the DES's films. Aimed at children aged thirteen and upwards six of them examine particular research projects in detail while the seventh and longest, *Talking of Science*, looks at the work of the four councils involved in the natural sciences. The films on the work of the Agricultural Research Council are the best of the short films, describing work on root growth and soil fauna at East Malling Research Station and techniques for growing virus-free plants to give higher yields. They not only describe important and valuable work lucidly, they are also very profes-

sionally made. The others have an amateurish air about them.

The Medical Research Council's film describes the work of the air pollution unit at St Bartholomew's Hospital, while studies at Edinburgh on how three- to five-year-old children develop the understanding of the meaning of words is the subject of the Social Science Research Council's film. Mapping the ocean floor with acoustic devices—with lots of animated diagrams—illustrates part of the work of the Natural Environment Research Council, while the Science Research Council alone plumps for a pure science project as the subject of its film, showing Sir Martin Ryle's work at the Cavendish Laboratory in Cambridge.

Talking of Science works its way across the whole spectrum of the natural sciences, lighting on each subject for an instant before rushing hurriedly on to the next. In this way the film manages to cram nuclear physics, molecular biology and genetics, virus diseases, plant pests, the rainfall and water cycle, marine biology, geophysics and the ionosphere and the troposphere into just thirty-three minutes, along with some spectacular leaps from subject to subject that leave the uninitiated gasping. Miss Buckton, an MRC geneticist, has been describing work on chromosome patterns and explaining the problems that an extra chromosome can cause in humans. The film cuts to Professor Sir Roy Markham in another laboratory who opens with the line "You've already heard about diseases caused in humans by defects in nucleic acid metabolism". Have we? Really? No one has actually mentioned the words before.

But if the films are not all well presented the content is all good, most of it easily intelligible and the scientists come across well. But the film makers might have been kinder to the scientists.

ENVIRONMENT

NATO's Clean Hands

THE Grand Committee Room of the House of Commons should be one of the most environmentally aware of chambers. Last Thursday, for the second time within a week it was subjected to details of how the European environment is being protected by the plethora of multi-initialled supranational European bodies. This time it was the turn of NATO. Dr Gunnar Randers, assistant secretary general of the organization and chairman of its committee for environmental challenges to modern society, addressed the behatted ladies and earnest schoolchildren of the Anglo-Norse Society on the subject of "Pollution", in particular outlining what the North Atlantic Treaty Organization is doing about it all.

Pollution control and NATO might seem odd bedfellows, but Dr Randers was at pains to point out that NATO is primarily a defensive organization rather than a purely military one, and he stressed the importance of the non-military aspects of its work. For 15 of its 25 years NATO has had a science programme which supports pure, non-military science. Five million dollars a year are spent on fellowships and conferences, and its recently established committee to study the environmental challenges to modern society is a logical extension of that work.

NATO's key advantages as a protector of the environment, Dr Randers said, are first that through its daily council meetings it has the ear of governments more directly than any other international organization and second that its approach is a highly pragmatic one.

Whenever any pollution problem is faced invariably the first thing that is discovered, according to Dr Randers, is that not enough is known about the subject and that further research is needed. But, he added, if everyone waits for full detailed information before acting, nothing will ever get done. Therefore, NATO sees its role not as a body to commission more research (that, said Dr Randers, is up to OECD) but as one that will act on present knowledge insufficient though it may be. Recommendations for action worked out by member nations are put to the NATO Committee on Environmental Challenges to Modern Society which considers implementing them. Dr Randers quoted the example of controlling oil spills in the North Sea, a problem that the International Marine Consultative Organization wrestled with for 15 years until NATO took it over a year ago and within a month produced an agreement that will prohibit oil spillage from 1975.

It is this sort of direct action in specific areas that makes NATO effective, according to Dr Randers. The problems of building safe cars to cut the carnage on the roads, of automating medical diagnosis to ease the strain on doctors, and of examining pilot plants for waste water disposal are other fields in which NATO is already working.

ENVIRONMENT

A Mixed Bag

THE half-day conference on the environment, organized jointly by *Nature* and the Royal Institution last week, covered, in a short time, problems which ranged from future trends in world population to pesticide pollution, with insights also into the available resources of fuels and metals and the problems associated with the gradual build up of carbon dioxide in the Earth's atmosphere.

Dr William Brass, reader in demography at the London School of Hygiene and Tropical Medicine pointed out, at the outset, that he sees no need for more people on Earth. He also sees hope that the current rapid increase in world population can be slowed down. In particular the decrease in fertility that has occurred in Japan and is now evident in Taiwan and Mauritius are indications that predicting population trends are unreliable. The decrease, in particular in Mauritius, is not the result of famine, family planning or even education said Dr Brass and it highlighted the fact that making forecasts of future population depends on inspired guesswork as much as anything. In this vein he pointed out that the recent publication *The Limits of Growth* (see *Nature* 236, 47; 1972) must not be considered as a prediction. According to Dr Brass

considering the MIT work in this light would be "scientific illiteracy".

Dr K. C. Dunham, director of the Institute of Geological Sciences, said that there is no great reason for pessimism "provided it is realised what is happening". He pointed out that the upward trend of the use of metals cannot continue for much longer. As time progresses ores of smaller and smaller enrichment are being mined and Dr Dunham pointed out that as far as the supplies of tin, lead and zinc were concerned, this cannot continue for there is a sharp break between the concentrated deposits of these ores and those of very low enrichment.

He called for greater reuse of metals and for more recycling. Synthetic substitutes would replace some metals but Dr Dunham does not think that a complete replacement is possible as there are "many operations in a sophisticated society which need metals".

Dr Kenneth Mellanby, Director of the Monks Wood Experimental Station, considered the vital question to be whether the benefits derived from the use of pesticides are greater than the disadvantages. He pointed out that it was essential to rely on pesticides to increase food production and that in fact if the use of all pesticides were banned then it would have the effect of reducing the population of the underdeveloped countries.

It is essential according to Dr Mellanby to ensure that the safest pesticides are used all over the world. Also Dr Mellanby stressed that a great responsibility lay at the door of the plant breeders to produce plant strains which are not susceptible to pests.

Dr Mellanby admitted that in some instances the use of pesticides had had deleterious effects but he was at pains to point out that there was no evidence that the amount of DDT in the oceans of the world now or in the future will ever reach such a level that it will prevent photosynthesis from occurring—and so alter the oxygen balance in the world.

The conference was led from oxygen balance to carbon dioxide balance by Dr J. W. Sawyer, director of research at the Meteorological Office. According to Dr Sawyer the direct effect of carbon dioxide on mankind is negligible with atmospheric content now being 319 parts per million to be compared with about 290 parts per million at the end of the nineteenth century. The indirect effect—the trapping of heat within the atmosphere, the so-called glasshouse effect—is however not so easily evaluated. The result is that the increase in average temperature of the Earth by such a mechanism is expected to be less than $\pm 2^\circ \text{C}$ —which is the average climatic variation since the last ice age which occurred 10,000 years ago. According

to Dr Sawyer, the problem of the glasshouse effect does not require further study but there is a need to discover where all the carbon dioxide that is released into the atmosphere goes to.

SOVIET SCIENCE

Antarctic Medicine

from our Soviet Correspondent

A NEW institute of medical research and practice has been established at the Molodezhnaya observatory—the "capital city" of Soviet Antarctic research. As with most Soviet projects, its aim is two-fold—the immediate implementation of current economic planning, and a somewhat more tacit programme of basic research.

When interviewed in *Pravda*, A. L. Matusov, head of the polar medicine department of the Arctic and Antarctic Scientific Research Institute in Leningrad stressed, first of all, the ever-growing significance of the polar regions as sources of ores, chemicals and fuels. If these regions are to be exploited, he said, the problems of polar medicine must be thoroughly investigated. So far such research has not been carried out on a full-time basis, but the time is now ripe for a continuous research programme. In particular he stressed that a detailed study of acclimatization is particularly important, especially an investigation of possible changes in the functioning of the internal organs and the nervous regulatory systems under very cold conditions. This work will be undertaken in the Antarctic, rather than in the Arctic, in part due to the more severe conditions of the Antarctic, and in part so that the new institute can also serve as a medical treatment centre for members of Soviet Antarctic expeditions.

The staff of the institute includes a psycho-physiologist, a surgeon and a therapist, and in addition to treatment rooms and research laboratories, the institute is equipped with no less than three computers.

Research into acclimatization, carried out at the institute, and at other Soviet research posts in the Antarctic, will include microbiological, hygienic and pathological factors, and the incidence rate of various illnesses. The susceptibility to disease and its dependence on age, profession, polar experience, time of year, and the geographical and climatological conditions of the individual Antarctic stations will also be studied. Finally, an investigation into psychosomatic factors is planned, including a study of the nervous system under various conditions, the efficiency of the sensory organs, and psychological compatibility within the small community of a research station.

NEW WORLD

Troubles Surface at Academy Meeting

by our Washington Correspondent

THE Annual meeting of the National Academy of Sciences last week once again provided an outlet for the troubles which have plagued that august body in recent years. The academy's leadership successfully blocked a move to give the membership much greater influence in the handling of classified work, but the manner in which this was done has caused deep resentment among some members which may yet backfire badly on the academy. At this meeting, the resignation of one member was finally accepted after a year's procrastination but that of another was deferred until the next meeting in November. A resolution calling for less emphasis on military force as an instrument of US foreign policy was accepted, and members voted to press ahead with a reorganization of the National Research Council—the academy's operating arm—without waiting for a solution to the deep and long-standing disagreements between the National Academy of Sciences and the National Academy of Engineering. Then, finally, academy members once again rejected an attempt by Professor William Shockley to persuade the organization to study the basis for his belief that blacks are genetically less intelligent than whites.

The academy's annual meeting is no longer the staid occasion of the past but a platform for dissent, partly at least because of the academy's role as an adviser to the government on science and technology. These activities, chiefly through the National Research Council, have grown to be worth \$35 million a year. They include controversial studies such as the effect of herbicides in Vietnam and the effect of automobile manufacturers to meet the standards specified by the Clean Air Act. But for some members of the academy, the acceptance of projects classified as secret has been a bitter pill. Members complain that since they are corporately responsible for the reports produced in the academy's name, the acceptance of classified studies makes them responsible for work which they cannot even see.

This is why Richard C. Lewontin, a geneticist from the University of Chicago, introduced resolutions at the academy's annual meetings in 1970 and 1971 calling for an end to classified work. The resolutions were soundly defeated, and a year ago Lewontin resigned from the academy. He was followed in November last year by Bruce Wallace, a geneticist from Cornell University.

Much of the confusion which has followed stems from a peculiar section of the academy's constitution, which requires that a resignation must be accepted by a meeting of the membership. Membership of the academy is almost as difficult to relinquish as to obtain. Lewontin's resignation was deferred at the meeting held last autumn but accepted last week, but Wallace will have to wait—and the establishment is hoping that he will reconsider in the light of other actions taken by last week's meeting.

One overall step towards Wallace's position was the adoption of a resolution to "ameliorate the difficulties and compromises with which the academy is confronted with respect to classified studies". The resolution was proposed by the president, Dr Philip Handler, and accepted by a lopsided margin. Immediately afterwards, Handler gave the impression that the resolution had united the membership and resolved many of the difficulties. Other reports of what went on behind the closed doors of the meeting suggest, however, that sweetness and light do not yet rule.

When Lewontin's resignation was deferred six months ago, Handler set up a committee on the handling of classified studies and, to avoid any suggestion of bias, appointed to the committee the youngest member of each of the academy's eighteen divisions. The committee's resolution was presented to last week's meeting and, independently, Wallace and Thomas Eisner, a neurobiologist, also of Cornell University, backed by eighteen other members, submitted their own resolution on classified research.

According to Handler's interpretation of the events, the committee's resolution was amended on the floor "because of certain awkwardnesses", the amendments being designed simply "to make what they wanted to do feasible". The amended motion, Handler said, was passed "virtually unanimously", although when asked later what the voting was, he said "this was unanimous". As for the motion submitted by Wallace and Eisner, Handler said "it was rescinded on the floor by Mr Eisner who said that this other accomplishes what we were trying to do. I think he says that it does not go quite as far as he would go, but it goes so much further than where we are that he is very pleased. . . . When we passed this other resolution he withdrew his."

Other members of the academy give a somewhat different account of what went on. According to these reports,

when the committee's resolution came up for debate, Dr Daniel S. Lehrman of Rutgers University, supported by Eisner, offered three amendments which in their opinion would have given the academy's members greater control over the acceptance of classified studies. The amendments would have caused the academy to reject studies which the council of the academy believes are needlessly classified and would have removed from the committee's resolution the provision that in cases of emergency the council could accept a classified study without first informing the membership. A third amendment would have given the membership, and not the executive, the final decision on whether or not to accept contentious classified studies. The first amendment was tabled (voted down without debate), and the other two were defeated by a margin of about two to one.

At that stage, Handler asked to be relieved of the chair, and read a resolution (see box) which he said he had drawn up the night before and which he hopes makes feasible what the committee's resolution was attempting to accomplish. After he had read his resolution, the only copy of which was in his hands, the proposers of the committee's resolution accepted it and it was passed by an overwhelming majority of the members present. Wallace's resignation then came up for debate, but it was agreed to suspend action on it until he had a chance to see the resolution that had just been passed.

The resolution submitted by Wallace and Eisner became the next item of business. In short, the resolution asked that unclassified synopses of classified projects should be submitted to the members on an annual basis, and called for a mechanism by which the membership could cancel contracts that it considered unacceptable. There were clearly many difficulties involved in this suggestion, because it might have led to the cancellation of contracts after they had been signed, but Eisner asked that a straw vote be taken on his resolution, the response to which from some members was "which resolution?" In view of the fact that the amendments to the committee's resolution had been voted down, he decided it was futile to press for a vote and decided then to withdraw his resolution.

Asked last week whether he is, as Handler suggested, happy with the final resolution, Eisner expressed surprise and annoyance that such an impression had been given. "I flatly deny that I am happy with the resolution," he said, "I

voted against it and I am personally very unhappy with it." At that time, Eisner had not even seen a written version of the resolution passed at the meeting, and he said "I shall have to wait for the official version of the resolution to reach me before I decide what action I shall take." Eisner also strongly denied that he had withdrawn his resolution because he was happy with the other one, and said "I believe that I was deliberately misrepresented."

Lehrman also said last week "I am not satisfied with the final resolution. I made a strong statement saying that the committee's resolution was weak and I voted against it." Lehrman said that he believes Handler's motion gained support from those who were relieved that it was not stronger, and from those who believed that it accomplishes what the committee intended.

Asked why he opposes the official version of the motion, Eisner said that apart from the fact that it contains the provisions that Lehrman's amendments sought to remove, it gives the council and not the membership the final decision on whether or not to accept a disputed contract. "To me," he said, "the critical thing about being a member of the academy or any other organization is that you delegate authority to the council for carrying out the members' wishes, but not the responsibility. Responsibility must always remain with individual members."

A more straightforward piece of business was the adoption of an antiwar resolution proposed by Dr Alexander Rich of MIT, which instructs the president of the academy to request that the President and Congress of the United States "evolve foreign policies in which the development and application of science and technology in industry, agriculture and health for the furtherance of human welfare are major elements, and reliance on military force, whether direct or indirect, is de-emphasized". Although Vietnam was not explicitly mentioned in the debate, the implication is clear.

The meeting also went through the formality of turning down a request by William Shockley, Nobel prizewinner for his work on transistors, that the academy conduct a study of the relationship of environment and genetic factors to intelligence. Shockley believes that genetic factors lead to a difference in intelligence between blacks and whites. Shockley's resolution was, however, defeated by a vote of 44 to 24, which is at least an improvement on his past attempts to steer the academy into seeming to support his views—he has been beating the same drum since 1966, but on every previous attempt, his resolutions have been tabled. He said last week that he is encouraged by the vote, and no doubt he will be at it again

in the autumn meeting of the academy.

The other piece of business concluded at the meeting last week was, according to Handler, the most important action taken. The members of the academy gave the council a mandate to press ahead with reorganization of the National Research Council along lines

that should make it more responsive to requests for multidisciplinary studies. "What it really has to do with", Handler said, "is whether or not the academy really intends to be an important element in shaping national policy in the future in the way in which we use science and technology".

Classified Studies

THE following resolution on the policy of the National Academy of Sciences with respect of classified studies was adopted at the Academy's annual meeting last week. The manner in which it was adopted is described in the accompanying article.

Preamble :

The role of the National Academy of Sciences, as an adviser to government, in some instances requires the Academy to concern itself with highly technical questions arising within the area of national defense. In some of the cases the Academy is requested to conduct its project/study on a classified basis. This is objectionable on two grounds. First, classification of the report of a study project runs counter to the tradition of open publication of scientific results. Second, classification of the conduct of the work places a barrier between Academy members and any project which might prove to be objectionable to some number of members, were they more fully informed of the details. The purpose of this resolution is to ameliorate the difficulties and compromises with which the Academy is confronted with respect to classified studies.

It Is Hereby Resolved That :

1. The Council of the Academy, in carrying out the responsibilities for contract approval assigned to it by Section II, paragraph II of the Academy bylaws, shall, to the maximum extent possible, insist that the projects and studies to be undertaken under these contracts shall be conducted on an unclassified basis. Insistence by an agency that a project or study be conducted on a classified basis shall, if a majority of the Council finds that classification is unnecessary, or that the proposed level of classification is excessive, constitute proper grounds for rejecting the contract.
2. Whenever the Council approves a contract providing for a classified project or study, the subject matter thereof shall forthwith be communicated to the Academy members in the following manner:
 - (a) An unclassified title and abstract adequately identifying the scope and intent of the work shall be submitted to the membership prior to the signing of the contract, provided, however, that this procedure need not be followed in cases deemed to be of special urgency by a majority of the Council. This abstract shall include indication of a deadline date for receipt of inquiries from the membership concerning further details of the contemplated project.
 - (b) More detailed information shall be available to any member on request, subject only to classification and proprietary restrictions.
 - (c) The contract shall include provision for issuance of an unclassified summary in addition to a classified report, whenever this is possible.
3. If prior to the deadline date, 10 or more Academy members, representing at least two institutions and two Sections of the Academy, agree that a research project approved by the Council is inappropriate for the Academy, special arrangements shall be made for presentation of their arguments to the Council. If the Council remains unpersuaded and proceeds with the project, as complete an account as possible of this matter shall promptly be submitted to the full Academy membership, for such action as the members deem appropriate.
4. The Council of the Academy shall develop, and inform the membership, a set of specific criteria and guidelines for acceptance of classified contracts by the Academy.

Set up in 1917 by President Wilson, the National Research Council is administered by the council of the academy; it is the organization which is responsible for carrying out the studies contracted by the academy. Organized as a mirror of the academy, the NRC has a number of divisions which roughly correspond to the chief scientific disciplines, and the chief thrust of the reorganizations proposed last week is designed to make the NRC more able to carry out multidisciplinary studies. In short, what is being proposed is that the NRC be reorganized into three assemblies, which will be concerned respectively with physical sciences, life sciences and social and behavioural sciences, and which would concern themselves with the welfare of their component disciplines. In addition to the assemblies, there will be set up a series of boards and commissions, each concerned with a continuing problem such as transportation, the environment or communication, and each spanning a number of scientific disciplines. It is suggested that the assemblies may provide a source of manpower for the various studies assigned to the boards and commissions, and that the whole organization would be governed by a board consisting in the main of the executive committee of the council of the National Academy of Sciences.

Another idea which may soon be brought into practice is to tackle some particularly tough problems (such as the study of the social, technical and economic implications of the water pollution control bill, that may be assigned to the academy by congress) with a committee staffed by postdoctoral students on a half-time basis. The suggestion is that the committee staff would do most of the donkey work for the committee members, in effect providing them with the basic information on which to make their recommendations.

Such a structure for the NRC was, in fact, proposed in 1969, shortly after Handler took over the presidency of the academy, but until now it has been bogged down by disagreements between the academy of sciences and the National Academy of Engineering. The basis of the disagreement is that although the National Research Council constitutes the operating arm of both the National Academy of Sciences and the National Academy of Engineering, responsibility for its governance is vested almost entirely in the hands of the NAS. The NAS has so far been reluctant to press ahead with reorganizing the National Research Council until it has settled its differences with the academy of engineering, but it now seems that it has been decided to go ahead regardless. Handler said last week that "the National Academy of Engineering is still in the house and I

hope it will stay in. There's no reason to think otherwise."

Whatever the result of the negotiations with the NAE, the National Academy of Sciences is clearly at a critical stage in its development. The challenges lying ahead are many, and its involvement in government decision making is clearly growing apace. It has a valuable role to play in the development of policies concerning science and technology, but its influence will depend in some large measure on the public recognition and acceptance that it can gain. That is why it is vital to settle the internal disagreements and differences of opinion over the handling of classified studies before the organization's public credibility is damaged. The next year or so is going to be a testing time for the academy in many respects.

AMERICAN PHYSICAL SOCIETY

Physicists in Protest

by our Washington Correspondent

ANOTHER venerable scientific organization, the American Physical Society, also saw its internal disagreements come to the surface last week. Like many other scientific societies, the APS has witnessed over the past few years growing activism from its younger members, who are concerned to change the organization from a learned society into a professional organization concerned with physicists and not just with physics. This movement has found its chief expression in an attempt to amend the APS constitution by grafting on to the present charter, which states "the object of the society shall be the advancement and diffusion of the knowledge of physics", the words "... in order to increase man's understanding of nature and to contribute to the enhancement of the quality of life for all people. The Society shall assist its members in the pursuit of these humane goals, and it shall shun those activities which are judged to contribute harmfully to the welfare of mankind".

Proposed by Robert H. March of the University of Wisconsin, and backed by about 275 other members of the society, the proposed constitutional amendment provided something of a talking point at the Spring meeting of the society, held in Washington last week, but it will not be put to a vote until the Autumn when members will ballot by mail.

The chief discussion of the amendment came in a debate, staged on the opening day of the meeting, which should have taken place between March and an opponent supporting the status quo. But in the event, the conservative wing of the association had a hard time finding a spokesman and March debated his amendment with Earl Callen of American University, a supporter of the intent of the amendment but not of its wording. Opposition to March's amend-

ment was based almost entirely on its attempt to steer the society into shunning those activities that are judged to be harmful to mankind.

Callen and several others have pointed out the obvious pitfalls inherent in such a suggestion. Its implication would clearly be to discipline or even expel those members who are judged to be doing "antisocial science," but the concept of a committee of the physical society sitting in judgment over a section of the membership is abhorrent to those who spoke against the proposal last week. And March's reply to his critics will do little to change their minds. Speaking in a press conference, March admitted that the wording of the amendment is unfortunate, but suggested that "there has been more heat than light in the way in which it has been interpreted". He said that he would not like to see the society draw up a code of ethics that the membership would have to follow, but saw the amendment as simply opening the way for discussion of many issues that are at present precluded by the restrictive way in which the society's executive committee interprets the constitution.

To support his case, March drew attention to a peculiar series of events surrounding the debut last week of the Forum on Physics and Society, a newly formed section of the American Physical Society whose objectives are summed up in its title. Formally established at the society's meeting held in January this year, the forum held its first symposium last week—a series of discussion papers on physicists and the Vietnam war. But although the papers presented at the symposium were judged by the organizing committee to be consistent with the forum's by-laws, the society's Executive secretary, Dr W. Havens of Cornell University, considered that since they deal with physicists and not with physics, they do not fit in with the society's constitution. He therefore decided that their abstracts should not be printed in the *Bulletin of the American Physical Society* with abstracts of other papers presented at the meeting.

March suggested that there is a damaging conflict when a legitimate section of the Physical Society sponsors a discussion which is ruled outside the terms of the society's constitution. He believes that it is incumbent on the society to provide a discussion of the ethical questions involved in undertaking research, for example, into weapons that are being used in the Vietnam war, but it became clear from his remarks that he is not happy himself about the amendment which bears his name. Asked whether he believes that it will be passed by the membership, March replied "I am sort of like George Wallace. I am quite confident that I am going to lose, but I am trying to send a message".

NEWS AND VIEWS

Molecular Mechanisms of Blood Clotting

ALTHOUGH the complex sequences of biochemical reactions involved in blood clotting are well known, there has been less agreement about proposals for a mechanism in terms of the three-dimensional interactions between molecules of known structure; much is known about what happens without understanding how. Progress towards bridging this gap between a biochemical and a molecular biological description of blood clotting is represented in the report by Tooney and Cohen on page 23 of this issue of *Nature*. By analysis of a modified fibrinogen which forms microcrystals, these authors conclude that the fibrinogen-fibrin conversion resembles a transition between two different polymorphic aggregates of monomers which incur no intra-molecular rearrangement.

Blood clotting is brought about when thrombin converts the soluble plasma protein fibrinogen into fibrin, which becomes insoluble by rapidly polymerizing into long fibres. The fibrinogen molecule consists of three pairs of identical chains of molecular weight of about 50,000, and the conversion to fibrin involves the cleavage of the small peptides from fibrinogen. Considerable uncertainty exists about the structure of the fibrinogen molecule. It was classified by Astbury with α -keratin, epidermin and myosin because it gave an X-ray diffraction pattern which indicated a molecular structure similar to these α -proteins. This α -protein structure is now known to take the form of two α -helices coiled about each other to form a two strand rope, but, unlike the simple rod shaped muscle proteins paramyosin and tropomyosin which are 100 per cent helical, fibrinogen is much less helical and thus a large fraction of the molecule is of unknown structure.

On the basis of various techniques conflicting proposals have been made that fibrinogen is a long thin molecule, that it consists of three globular portions linked by two helical rods, that it has an open spherical lattice structure, that the fibrino-peptides are close together or alternatively far apart. In electron micrographs obtained by shadow casting the fibrinogen molecule appears to be about 475 Å long, whereas preparations of fibrin have an axial periodicity of 225 Å; the conflicting explanations involve shrinkage of the molecules or staggering the molecules by half their length axially. Although Tooney and Cohen's report does not solve all of these problems, considerable light is thrown on the likely molecular mechanisms involved in clotting.

Tooney and Cohen have discovered that when fibrinogen is treated with certain bacterial proteases, part of the molecule is cleaved to yield fibrinogen Ps which is still convertible to fibrin by thrombin; the heaviest of the three chains comprising the molecule is affected. At high ionic strength fibrinogen, fibrinogen Ps and fibrin can all yield similar structures with axial periodicities of 225 Å. At lower ionic strength fibrinogen forms fibres with no visible periodicity in the electron microscope and fibrinogen Ps precipitates as highly ordered rod-shaped patterns, termed as obliquely striated and orthogonal sheet, both with transverse bands separated axially by

450 Å. Because alternate bands are shifted laterally with respect to each other a true axial repeat of 900 Å is generated in both cases. The authors infer head to tail aggregation of molecules 450 Å long. Furthermore, in view of the similarity between the high ionic strength structures given by fibrin Ps (that is, fibrinogen Ps precipitated with thrombin) and native fibrin they infer close similarity between the molecular structures of fibrinogen and fibrinogen Ps.

Perhaps the most interesting observation of Tooney and Cohen, however, is that superposition of variously translated images of the orthogonal sheet structure can produce a number of patterns similar to some intermediate patterns observed and, in particular, to the native fibrin pattern with a period of 225 Å. This suggests that there may be one fundamental array, say the orthogonal sheet, which by aggregating in specific modes can generate various structures. Thus fibrin, say Tooney and Cohen, may be a polymorphic form of fibrinogen, and the transition between the two would be accompanied by a minimal intra-molecular rearrangement.

The fact that a modified fibrinogen can adopt a regular microcrystalline form holds out promise that further details of the molecular structure and its possible interactions will be determined so that theories on the molecular mechanism of blood clotting can be tested.—
From a Correspondent.

T and B Cell Cooperation

IMMUNOBIOLOGY is in the middle of a storm and the confusion is considerable. Three articles in the current issue of *Nature New Biology* (May 3) all exemplify what is happening; they all concern in one way or another the cooperation between T and B cells, a theme which is currently of great interest is cellular immunology.

Schimpl and Wecker (*Nature New Biology*, **237**, 15; 1972) studied the response of mouse spleen cells to stimulation by sheep red blood cells (SRBC) *in vitro*. In their first experiments they established that T cells were a prerequisite for an immune response measurable as 19S plaque forming cells. They did this by showing that spleen cells treated before culture with anti- θ antisera were unresponsive, but they found that the supernatant from simple spleen cultures could occasionally make an inadequately anti- θ treated cell population respond quite well to SRBC. These supernatants were always inactive on spleen cells that had been rigorously treated with anti-theta. Schimpl and Wecker therefore conclude that spleen cell populations *in vitro* can liberate a T-cell expanding factor (TEF) which is perhaps similar in its effect to the twin stranded RNA which Cone and Johnson (*J. Exp. Med.*, **133**, 665; 1971) have shown can expand the influence of thymocytes *in vivo*.

Schimpl and Wecker went on to experiment with supernatants from mixed cultures in which spleens from two different strains of mice had been growing for twenty-

four hours. These supernatants, added to cultures which had no T cells, facilitated a response to SRBC as good as if all the T cells had been present. The remarkable agent responsible is termed the T cell replacement factor (TRF). Schimpl and Wecker present evidence that TRF is a T cell product and conclude that such materials may well be active *in vivo* during cooperation between T and B cells.

Feldmann and Basten's experiments (*Nature New Biology*, **237**, 13; 1972) are not dissimilar to those of Schimpl and Wecker. Irradiated CBA mice were injected with syngeneic thymocytes and keyhole limpet haemocyanin (KLH), and some days later the activated thymus cells (previously termed educated) were recovered from the spleen. These cells are one constituent of their eventual mixed spleen cell cultures; the other component derived from the spleens of mice which had been immunized with dinitrophenylated flagella (DNP-Fla) from *Salmonella adelaide*.

Initially both spleen cell populations were brought together in culture in the presence of DNP-KLH and a vigorous anti-DNP response was recorded. The response is brought about because the carrier primed (T) cells respond to the KLH and promote the response of the DNP-Fla primed (B) cells. Then carrier and hapten primed cells were placed in two compartments separated by a nucleopore membrane of 1μ porosity. The anti-DNP response was still observed in the chamber carrying the hapten primed B cells. Elimination of T cells from the DNP-Fla primed spleen population did not affect the outcome. Feldmann and Basten conclude that a soluble factor produced by the primed T cells responding to KLH diffuses through the nucleopore membrane and promotes the response of the B cells. Furthermore they conclude that the factor is specific in that antigens other than KLH failed to elicit its production. It must be mentioned that Feldmann and Basten conducted their cultures in the presence of SERF—a material sometimes found in foetal calf serum which is capable of enhancing the response to SRBC of neonatally thymectomized mouse spleen cells *in vitro*—and they do not comment on the role of this factor.

The problem was attacked from a different angle by Wilson and Feldmann (*ibid.*, **237**, 3; 1972). They were concerned with the binding to B cells of DNP conjugated to various carriers. They immunized mice with DNP—F γ G (fowl γ globulin), and the spleens of these animals were subsequently collected and shown to contain cells which would form rosettes with DNP-SRBC. Furthermore, most of the rosette forming cells were not inhibitable in their capacity to form rosettes by treatment with anti- θ antisera and they could therefore be thought of as B cells. These cells were incubated with DNP conjugated to lysine, as a monovalent form, to HGG, twelve molecules of DNP/protein molecule, or to flagellin, with many DNP units per molecule of protein. The recovery of the capacity to form rosettes with DNP-SRBC was determined after incubation with the various hapten-protein conjugates. Wilson and Feldmann found that DNP-lysine could inhibit rosette formation but that recovery was rapid either at 4° C or 37° C. DNP₁₂HGG caused inhibition from which recovery was negligible after 4½ hours at 4° C but rapid at 37° C. The inhibition brought about by DNP-Fla was pronounced and had a half-life of about 9 hours even at 37° C. It

was shown further that the capacity to inhibit rosette formation was directly related to immunogenicity in that good inhibitors were strongly immunogenic.

Wilson and Feldmann, satisfied with what they regard as a good correlation between binding capacity and immunogenicity go on to conclude that the ability of polymeric antigens (at least *in vitro*) to immunize B cells with the normal helper mechanisms is related to their cell-surface binding potentiality. They envisage that T cell collaboration is to be thought of as providing a more rigid framework for the carrier molecule, mimicking the structure of T-cell independent antigens and enabling the hapten to remain in contact with B cell receptors for a sufficient period of time to immunize.

All these three systems of experimentation are *in vitro*, all measure the γ M rather than the γ G component of the immune response. There will follow a difficult period of reconciliation before the *in vivo* mechanism or even the significance of cooperation between T and B cells can be established. At present it must be regarded as an interesting phenomenon which provides intellectual exercise for many.—From a Correspondent.

EARTH SCIENCES

Rifts, Ruptures and Continental Drift

from a Correspondent

CONTINENTAL drift, seafloor spreading and plate tectonics have become the trinity of the earth sciences, and the NATO Advanced Study Institute which met recently (April 10–14) at the University of Newcastle upon Tyne to consider the implications of the earth sciences, provided some of the details of the creed over a wide range of subjects. Participants were asked in effect to take the case for drift as proven in principle and to examine how the concept might have a bearing on their subjects.

Contributions on palaeomagnetism set the scene for later subjects, but also served to emphasize the continuing need for more precise information on past reconstructions. Subsequent proceedings during the week bore this out, but a clear demarcation of interests was discernible. Although many earth scientists concentrate on post-Palaeozoic reconstructions, there is greater scope for speculation about the preceding time span which is very much longer. By tackling this directly through palaeomagnetism, or indirectly by ap-

plying principles founded on current and post-Palaeozoic events, ideas relating to one of the more important underlying questions of the meeting are emerging. Has drift always operated in the same way, or has it undergone changes during the Earth's history. In other words, to what extent is it legitimate to indulge in palaeo-plate-tectonics?

Dr E. Irving (Geomagnetic Laboratory, Canada) and colleagues recognize five periods of orogenesis since the Archaean (2,500 m.y.) of the Precambrian Canadian Shield, marked by sharp directional changes in the polar wander curves. Implied compounding of previously distinct continental units was also inferred by Dr M. W. McElhinny (Australian National University) from new Soviet palaeomagnetic data for Eurasia in the pre-Mesozoic. Dr E. A. Hailwood (University of Southampton) related the Hercynian fold belt in Europe to the closing of a proto-Atlantic. Using the evidence of seafloor spreading rates, Dr P. J. Coney (Middlebury Col-

lege, Vermont) concluded that the North American cordillera was developed by deformation of the North American plate as it passed westward over an eastward dipping subduction zone, rather than as a direct product of subduction. How then might subduction and continental sutures in the pre-Mesozoic be accurately recognized? Professor E. Moores (University of California) examined implications for petrology to this end by presenting a comprehensive review and analysis of Alpine peridotite types. He also noted their absence from middle and early Precambrian deformed regions, so inferring a secular change in the nature of the oceanic lithosphere. On more general considerations, Dr W. M. Cady (US Geological Survey) felt that it would be unwise to extrapolate uncritically from more recent manifestations of subduction.

On the subject of rifting, the search continues for evolutionary relationships between relatively small scale swells and basins, major rifts and continental ruptures. Professor K. Burke (University of Toronto) identified eleven regions of dome uplift in Africa capped by three-armed graben structures. These can, but do not necessarily, evolve into full scale continental breaks depending on how they are influenced by contemporaneous plate movements. Professor Burke believes the swells are isostatic responses to partial melting at the base of the lithosphere. Professor H. Faure (University of Paris), however, showed how swells and basins can develop over undulations of the asthenosphere, and their developmental history related to differential lateral movement of lithosphere over them. Hot spots, which were frequently discussed, give rise to oceanic volcanic island chains in a geometrically analogous fashion. There is evidently continued scope for considering the effects of plates over mantle irregularities and inhomogeneities.

The need for more attention to deeper layers of the Earth was implied by Professor K. M. Creer (University of Newcastle upon Tyne), who, in discussing an improved method of determining pre-Mesozoic continental movements, pointed out that a better knowledge is required of the behaviour of the Earth's magnetic field. Professor S. K. Runcorn (University of Newcastle upon Tyne) emphasized that plate tectonics was incomplete without a plausible mechanism, and this must reside in the mantle.

It is evidently easier to find implications of continental drift in those branches of earth science which are concerned with structure and fabric, such as tectonics and petrology, than it is in those subjects where potential links of ideas are inevitably more circuitous. Thus implications for palaeontology, sedimentology and geomorph-

ology (the last two of which were scarcely mentioned at Newcastle) are essentially dependent in whole or in part on the intermediate implications for palaeoclimatology and palaeogeography. Professor A. E. M. Nairn (Case Western Reserve University) illustrated this by suggesting that at times of mid-ocean ridge development, there is widespread spilling over of oceanic waters on to continental margins, giving such marked universal transgressions as that of the Cenomanian (about 120 m.y.), and also causing provincialism in faunas which were previously homogeneous.

Dr A. Hallam (University of Oxford) considered the complex relationships between environmental changes brought about by continental drift and their effect on invertebrate evolution, diversity and extinctions. The need for palaeontologists to know more about the conditions in which past organisms lived is being met by the increasing refinements of palaeoclimatology. Dr P. Robinson (University College, London) showed how past climates might be predicted by superimposing Köppen's scheme on past continental shapes and configurations. Dr L. A. Frakes (Florida State University) gave a glimpse into the potential for palaeo-oceanography by postulating a Tertiary Indopacific, which, freed from Indonesian barriers, would have had a single circulatory pattern. The extent of this would have led to greater warming of waters passing along the equatorial zone to give higher seawater temperatures in the region of the present western Indian Ocean. Professor N. D. Watkins (University of Rhode Island) produced a palaeo-oceanographic model of waters between Australia and Antarctica on the evidence of post-Mesozoic seafloor sediment cores. In the contribution of Professor R. W. Fairbridge (Columbia University), there was a cross-over of several subjects of the meeting, as he drew tectonics and climatology together on the problem of glaciations. He believes that each influences the other by complex feedback.

The various tectonic and allied implications of continental drift were taken up again in the closing contributions. Professor A. M. Goodwin (University of Toronto) inferred that some form of plate tectonics has been operating since about 3100 m.y., though the mantle has undergone compositional changes over this period. Professor J. Sutton (Imperial College, London) envisaged that before 2,600 m.y., the crust was notable more for its overall mobility than for the presence of stable blocks. Once such blocks had evolved, they were subjected to periodic world wide phases of mobility, though he had reservations as to whether true oceans

were formed until the late Precambrian. Dr P. M. Hurley (Massachusetts Institute of Technology) weighed the evidence for and against subduction in the Precambrian and concluded that such extrapolation was valid, though he admitted to having just changed his mind. Professor R. M. Shackleton (University of Leeds) took the opposite view and presented a striking example of ancient lineaments in southern Africa which are aligned across the later Mozambique orogenic belt. He suggested the term "hallucinosutures" for such features.

REPRESSORS

Translational Restarts

from our Cell Biology Correspondent

ANYBODY who is feeling jaundiced and thoroughly satiated with molecular biology should, if they are at all interested in regaining their appetite for the subject, find themselves a secluded and comfortable corner in a library and spend half an hour or so savouring the article by Platt, Weber, Ganem and Miller in the current issue of the *Proceedings of the National Academy of Sciences* (69, 897; 1972). It is an example of first rate craftsmanship; a story of sophisticated experiments flawlessly carried out. The geneticists of the group have isolated a set of fifty odd amber mutants in the *i* gene of the *lac* operon of *Escherichia coli*, the structural gene for the subunit polypeptide of the tetrameric *lac* repressor protein. These mutants, at various positions in the gene, cause the premature termination of translation of *i* gene messenger RNA and so give rise to truncated *lac* repressor polypeptide. Five of these amber mutants map near the 5' (amino) terminal end of the *i* gene—the genetic evidence for this conclusion is apparently to be reported elsewhere—and should give rise to only a very short non-functional fragment of the repressor protein. But in diploid strains these five mutants negatively complement with the wild type *i* gene. In other words in addition to the short amino-terminal fragment of the repressor polypeptide these five mutants also specify a much larger fragment which can associate in diploid strains with intact wild type repressor subunits to give rise to a mixed, mutant and wild type, tetrameric repressor protein. Presumably, therefore, in these early amber mutant strains the translation of the *i* gene mRNA is reinitiated and completed after the early termination event dictated by the mutant amber codon. There are, of course, precedents in other genes for such transitional restarts.

To prove this point Platt *et al.* engineered a temperature inducible prophage, λ h80 *dlac*, which carries an *i*Q

promoter and an i^{100} early amber mutant gene. This prophage overproduces repressor some two hundred-fold which means sufficient of the i^{100} repressor protein can be obtained for its amino-acid sequence to be determined and its properties characterized. The tetrameric repressor specified by the i^{100} gene binds the inducer IPTG; it cross-reacts with antibody against wild type repressor albeit to a ten-fold lesser extent than the wild type protein, and it has a molecular weight of about 135,000 which is about 8 per cent less than the wild type repressor.

Analysis of the monomeric subunits of the i^{100} repressor indicates that the polypeptide chains have a molecular weight of about 34,000 and the amino-terminal sequence is Ala-Glx-Leu-Aspx-. This same sequence occurs at residues 43-46 of the wild type subunit and in the wild type subunit it is immediately preceded by a methionine residue at position 42. This is the first methionine residue in the wild type sequence. Methionine is, of course, in a formylated state the initiating amino-acid of bacterial proteins and it seems clear that the amber mutation in the i^{100} strain precedes position 42 and that after translation is terminated by the amber codon it reinitiates at the methionine codon at position 42, goes to completion and then the initiating formyl-methionine residue is cleaved to leave the wild type position 43 amino-acid as the amino-terminus of the i^{100} repressor subunit. Although the position 42 methionine codon can act as an initiation signal in these circumstances, at least 99 per cent of the repressor subunit chains specified by the wild type i gene are of full length with an internal methionine residue at position 42. What tells the translation machinery to read the position 42 codon as a start signal after premature termination is a fascinating and unsolved question.

Finally, although the i^{100} repressor protein binds IPTG and is a tetramer it does not *in vivo* repress the *lac* operon. Platt *et al.* therefore conclude that the first forty-two amino-acids of the repressor subunits are essential for repressor activity, either because they are directly involved in association with the operator or because they are required if the repressor is to have the conformation it needs to recognize the operator. And as Jobe, Riggs and Bourgeois (*J. Mol. Biol.*, **64**, 181; 1972) have stressed, allosteric mutants of *lac* repressor protein have strikingly different affinities for inducer and the *lac* operator. Indeed, as they comment, repressors from different inducible strains of *E. coli* which have hitherto been thought of as wild type and identical in fact differ considerably in their kinetics on binding to inducer and operator.

CELL BIOLOGY

Aspects of Ageing

from a Correspondent

Two principal approaches to the problem of cellular aspects of ageing emerged at a joint meeting of the British Society for Cell Biology and the European Cell Biology Organization Study Group on Ageing at the University of East Anglia on April 5-7. First, that ageing is a clonal phenomenon resulting from the accumulation of errors at the molecular level, and, second, that ageing is a final and deliberate result of cell differentiation and is essentially an intercellular process.

The Searle lecturer, Dr L. Orgel (Salk Institute, La Jolla), reviewed the molecular theories of ageing, but concluded that no single theory is likely to prove correct. Rather, cellular ageing is probably the result of an accumulation of errors of various kinds, leading to the interaction of levels of imperfection, the breakdown in any system being both caused by and the cause of breakdowns in other systems. In a critical review, Dr H. von Hahn (Neurologisches Universitätsklinik, Basle) pointed out that research on nucleic acids and ageing had been too heterogeneous, inconclusive, contradictory and non-reproducible to be of much value, and hoped that new ideas would emerge before the next ECBO meeting on the subject. Dr J. Gallant (University of Washington, Seattle) described a bacterial system in which errors in protein synthesis could be elicited, their frequency measured and their effect on survival assessed, and reported that present evidence suggested that bacterial cells can tolerate a great deal of aberrant protein.

Dr I. Gibson (University of East Anglia) said that ciliate protozoans undergo a fission-dependent series of phenotypic changes demonstrable in terms of sexual immaturity, maturity and senescence, and controlled by the macronucleus.

Several speakers considered the implications of Hayflick's theory that normal somatic cells inevitably age and die after a finite number of replications. Dr R. Holliday (National Institute for Medical Research, Mill Hill) considered evidence for the accumulation of defective proteins and reported that a minor fraction of heat-labile G6PD and 6PGD appears as diploid cultured human fibroblasts become senescent, but that this fraction represents 20-30 per cent of the total activity just before death. Dr A. Maciera-Coelho (Villejuif) reported that chick fibroblasts given 100 rads at intervals show accelerated growth decline in proportion to the total irradiation received. Dr Y. Courtois (Institut St Pierre, Paris) considered that changes in chromatin in ageing chick fibroblasts might be associated with an imbalance in nucleoprotein synthesis. Mr P. Godsell and Dr M. Balls (University of East Anglia) doubted whether Hayflick's theory could be applied to amphibian cells, because a number of stable diploid and haploid lines from a number of species had been maintained *in vitro* for more than 200 cell generations. Drs A. Guillozo, G. Gueguen and Y. Le Guilly (INSERM, Rennes) described morphological, ultrastructural and metabolic changes in ageing diploid human liver cell strains similar to the changes occurring in senescent animals.

Professor H. Woolhouse (University of Leeds) said that, if plant cells age at all (they seem to carry on indefinitely

Origin of X-ray Background Radiation

CAN the X-ray background radiation of the universe be explained in terms of the smoothed out radiation from a number of discrete sources? This question has yet to be resolved to the satisfaction of all astronomers, but in next Monday's *Nature Physical Science* (May 8), A. C. Fabian lays down the framework within which any satisfactory resolution of this problem must lie. Fabian concludes, on the basis of recent observations, that it is still not possible to rule out the idea that this radiation originates within clusters of galaxies; but it does seem clear that, as pointed out by Wolfe and Burbidge (*Nature*, **228**, 1170; 1970), an origin in sources equivalent to superclusters is not consistent with the observations.

Determination of the nature of the background remains, of course, largely a matter for statistics to resolve, and what Fabian has done is to provide the

appropriate statistical techniques in a form immediately suitable for the problem. It will be interesting to see how future observations can be analysed in this way; at present, the data allow a model in which the X-ray background originates in clusters which have a Gaussian distribution and are characterized by a radial size less than 3.5 Mpc. The space density of cluster centres in such a model would be about 10^{-3} Mpc⁻³ to produce the observed flux.

According to Fabian, it is a relatively simple matter to allow cluster parameters (size, density and luminosity) to vary with redshift when determining the overall background produced in models of this kind. It seems, therefore, that as observations improve and more data are gathered the study of the X-ray background will provide an invaluable tool for the study of the structure of the universe.

in tissue culture), it is in the context of the whole organism; he then reviewed the eleven real facts about plant ageing discovered in the past 85 years. Professor T. Villiers (University of Natal) outlined his intriguing new theory of seed ageing, which states that seeds survive in the soil for long periods not because they are completely dormant, but because they are metabolically active and can carry out essential repair and maintenance.

Echoing von Hahn's scepticism, Dr P. Wilson (Imperial Cancer Research Fund, London) suggested that previous work on quantitative changes in enzymatic activity in ageing tissues had been confusing and unprofitable. Her studies on thirteen enzymes in kidney, lung and liver from C57 mice in four age groups had shown only that (at least in mice) sex modifies the effects of ageing, and that some changes probably occur in mitochondrial respiratory enzymes. Dr Wilson and her colleague Dr C. Rowlatt agreed that ageing in tissues is initially a focal process and, like many other participants, felt that various cells in various organs probably aged in different ways, even in different individuals of the same species.

Dr L. Robert (Centre National de la Recherche Scientifique, Paris) considered that ageing in extracellular materials is caused by two separate phenomena—a continuous shift in the proportions of the various extracellular molecules synthesized, and the modification of the extracellular matrix with age, with feedback affecting the metabolism of the cells themselves. Dr U. Schmidt (University Charity Clinic, Berlin) also felt that ageing is not merely a process of fibrosis or sclerosis, but the result of variations in rates of synthesis and degradation. Dr P. Leuenberger (University Eye Clinic, Geneva) described an age-correlated thickening of the capillary basement membrane in the retinas of spiny mice and rats, and suggested that similar changes in pathological conditions, such as diabetes, might be caused by the acceleration of a normal process rather than by the induction of new phenomena. Dr W. de Priester (University of Leiden) described the accumulation of dense lysosomal bodies in the midgut epithelium of the fly, *Calliphora*, and pointed out that insects are of great value in ageing studies because there is virtually no cell renewal in the adult.

Mr A. Payne (University of Edinburgh) discussed the progressive decline with age in the ability of mice to produce specific antibody. Because young and old cells are equivalent in their ability to mount a response in young irradiated hosts, and because young cells cannot significantly improve the response of old irradiated recipients, he considered that this is an organismal

phenomenon caused by some cellular, tissue structural or hormonal deficiency. Professor D. Bellamy (University of Cardiff) discussed the thymus as a model for studies on ageing and concluded that cortisol-induced involution of the thymus might represent the acceleration of a normal process—by accelerating cell death without inducing a corresponding increase in cell proliferation.

The overall impression remaining from this conference is, first, that there is no general agreement on what ageing really is and, second, that it is difficult to make comparisons between aged and non-aged cells, tissues or organisms. Are the aged a physiological elite, survivors of the functional failures or parasitic infections which normally kill at an earlier stage? Does ageing occur in the absence of disease? Dr Schmidt surprised the audience with his report that all 3,000 "healthy" over-65s examined in the ageing study had two to five diseases, many of which were treated successfully (so some good came out of the work). Dr Robert made the one irrefutable point—that at each meeting when he saw his colleagues in the ageing field again, he felt that they seemed a little older than at the previous conference.

SOLAR MAGNETISM

Sunspot Variations

by our Cosmology Correspondent

VARIATIONS in the magnetic field of the Sun may be explained in terms of the dispersion of sunspot groups over the solar surface. J. Tuominen, of the University of Helsinki, has now presented evidence, in the form of sunspot counts, which supports his theory that regular changes in the magnetic field of the Sun occur because the following part of a bipolar sunspot group always disperses more rapidly than the leading part (*Astrophys. Lett.*, **10**, 175; 1972).

The observations which led to this suggestion were made by Severny (*Nature*, **224**, 53; 1970), who found that the intensity curve of the solar magnetic field is similar to that of magnetic stars, and in particular that the Sun behaves like a quadrupole magnetic rotator. Tuominen pointed out that because of the rapid dispersion of the following part of sunspot groups there must be a widely distributed general magnetic flux balancing the strong localized field in the leading parts, and that variations associated with the sunspot cycle could explain Severny's observations (*Nature*, **228**, 1179; 1972).

Regulation of the Tryptophan Operon

THE set of five contiguous genes which specify enzymes involved in the biosynthesis of tryptophan in the *Escherichia coli* chromosome constitute an operon for the expression of these genes (designated *Operator*, *trpE*, *D*, *C*, *B*, *A*) is coordinately controlled by the intracellular concentration of tryptophan. When this amino-acid is present the genes are switched off; conversely when the cells are starved for tryptophan this set of genes is expressed. Moreover, as several groups have shown, tryptophan seems to exert its regulatory effect by first interacting with the product of a regulatory gene *trpR* which is not adjacent to the operon.

Because the transcription of the *trp* operon starts near the beginning of the *E* gene and for other genetic reasons it has often been suggested that the polypeptide chain specified by this first structural gene of the operon also plays a part in the feedback control of the entire operon. The latest experiments of Hiraga and Yanofsky, which are reported in next Wednesday's *Nature New Biology* (May 10) indicate, however, that neither intact *E* gene messenger RNA nor intact *E* gene polypeptide are essential for the regulated expression of the other four genes (*trpD*, *C*, *B*, *A*) of the operon.

Hiraga and Yanofsky have reached

this somewhat surprising conclusion from analysis of a deletion mutant strain designated $\Delta trpE5$. In this strain, in spite of the absence of a complete *trpE* gene, the expression of the genes *trpD*, *C*, *B*, *A* is repressed when tryptophan is added and derepressed when tryptophan is removed from the culture medium. In other words, in spite of lacking at least part of the *E* gene the remainder of the operon in cells of this strain is sensitive to feedback regulation.

But just how much of the *trpE* gene is missing? By comparing the time taken to transcribe the *trp* operon in a strain in which the operon is intact with the time taken in the strain carrying the $\Delta trpE5$ deletion, Hiraga and Yanofsky have shown that it takes about 50 seconds longer to transcribe the intact operon than to transcribe the deleted operon. If transcriptase polymerizes RNA at a rate of about thirty-three bases a second some 1,650 bases of the *trpE* gene must be missing and the entire gene comprises only about 1,700 base pairs. In short, 97 per cent of the *trpE* gene seems to be deleted in $\Delta trpE5$, but the other genes of the operon are still subject to feedback control. It is difficult therefore to avoid the conclusion that this regulation does not depend upon the production of an intact *E* gene messenger.

The new evidence which supports this idea comes from a study of sunspot variations from 1966 to 1969. In each of the four years (that is, every thirteen Carrington rotations) the number of spots varies in a way which is clearly related to the magnetic field variations observed by Severny at different latitudes. According to Tuominen, about half of the total solar magnetic field of ~ 1 gauss is related to the activity of visual sunspot leaders.

NUCLEOTIDE SEQUENCES

Bigger and Better

from our Molecular Biology Correspondent

THE determination of ever longer tracts of nucleotide sequences of viral and ribosomal RNAs has created as many questions as it has answered. The most striking achievements so far have centred on the RNA of the small phages of the R17 group. Adams, Spahr and Cory (*Biochemistry*, **11**, 976; 1972) have now consolidated and extended the sequence at the 5' end of R17 RNA so as to locate precisely the beginning of the first protein cistron, which occurs after a long leader sequence of unknown purport.

The R17 RNA has a built-in end-group label, in the form of a triphosphate group on the terminal guanosine residue, and 5' terminal fragments in a limited nuclease digest can therefore be identified by the appearance of a unique spot in electrophoresis patterns of total hydrolysates of the isolated components. Four such terminal fragments were in fact found in a mild ribonuclease IV digest, the longest of which proved to contain 117 residues. This is the tract for which Adams *et al.* now give the complete sequence, and it turns out to be just sufficient to establish the overlap with the fragment containing the start of the A-protein cistron, which was fully sequenced earlier. The first seven residues of the latter coincide with the last eight of the 117-residue terminal fragment.

In view of the vanishing low probability that a given sequence of eight nucleotides will appear twice in an RNA of moderate length, and the reported finding of an overlapping piece by the Cambridge group, there thus seems no doubt that the first cistron begins with an initiation codon, starting at position 130. This is precisely the result obtained for the RNA of the closely related phage, MS2, by Fiers's group in Belgium who a few months ago completed the sequence of the first 125 residues (and for another member of the same family, $\phi 2$, the first seventy-four residues at least are also homologous). With a few dubious looking bulges, a base-pairing scheme of sorts can be constructed by

making a hairpin of the sequence, but the longest unbroken Watson-Crick sequence is only five base pairs, and one would therefore expect its stability to be relatively marginal.

The purpose of the long leader sequence in these RNAs remains an absorbing mystery. That such strict homology is maintained in variants of the phage isolated in different parts of the world suggests a specific functional significance. The essentially unrelated phage, Q β , also has a long untranslated sequence at its 5' end, and within this Adams and Cory found two sizable segments that had closely similar counterparts in the same region in R17. Adams *et al.* feel that it may be involved in replication, providing, for instance, polymerase initiation sites. Another striking feature is that four initiation codons occur in the leader region, but are evidently in themselves insufficient to attract the ribosome. Other as yet unrecognized signals must therefore be implicated in the initiation process.

On the ribosomal RNA front, an important advance comes from Santer and Santer (*FEBS Lett.*, **21**, 311; 1972), who have sequenced both a twenty-six-nucleotide fragment split from the 3' end of *Escherichia coli* 16S RNA by T₁ ribonuclease, and the fifty-nucleotide fragment specifically knocked off the 3' end *in situ* by colicin E₃. The terminal segment has some curious features, most of all the remarkable density of methylation in this region. In an RNA containing few methyl groups, it is surprising to find no less than five within

a group of a few nucleotides, four of them in two adjacent residues, a dimethyladenine and a dimethylguanine. There is an antibiotic, kasugamycin, that evidently recognizes this sequence—which, as the lethal effect of colicin E₃ also demonstrates, is vital for ribosomal activity. Dahlberg and his colleagues showed last year that in a mutant devoid of the two dimethylated bases kasugamycin is without effect. The resistant strain is thought to lack a methylase which presumably recognizes a tract of sequence in this region.

The action of colicin E₃ on the ribosome is, in fact, a rather complex matter. The removal of the terminal nucleotide segment *in vitro* was established by work from Nomura's laboratory, and also by Boon, who now gives some further details (*Proc. US Nat. Acad. Sci.*, **69**, 549; 1972). A curious feature is that the colicin will not work on the 30S subunits alone in the absence of the larger subunit. When the 50S subunits are added back, the colicin at once takes effect. The agent of cleavage is not the 50S subunit after some form of sensitization by the colicin, for pretreatment of either subunit with colicin produces no effect. Some strains of *E. coli*, especially those that themselves synthesize colicin E₃, are resistant, and are known to carry an "immunity factor". Boon has now carried out titrations with preparations of the immunity factor which show that this acts by binding to the colicin, rather than to the ribosomes. Once the colicin concentration exceeds that of the factor, the

Infectious DNA Proviruses

IF it is assumed that the genetic information of RNA cancer viruses persists in infected cells as a double stranded DNA molecule, the provirus, which is synthesized by viral reverse transcriptase using the single-stranded genomic RNA of the virion as a template, it should be possible to isolate the proviral DNA from transformed cells and perhaps use it to infect and transform other cells. Such an experiment is undeniably ambitious, but according to the report of Hill and Hillova, which is published in next Wednesday's *Nature New Biology* (May 10), it can be done.

Hill and Hillova extracted DNA from rat XC cells transformed by the Prague strain of Rous sarcoma virus (PR.RSV) and from hamster cells transformed by a temperature sensitive mutant strain of Schmidt Rupp Rous sarcoma virus (SR.RSV) and purified the DNA by centrifugation in caesium chloride gradients. They then repeatedly exposed cultures of fresh chick fibroblasts, previously washed in DEAE-dextran solutions, to aliquots of the

DNA from the transformed cells and searched for foci of transformed chick cells and for evidence of the production of progeny PR.RSV or SR.RSV particles. As controls they exposed similar cultures of chick cells either to DNA from transformed rodent cells which had been digested with DNAase or to DNA from calf thymus.

As optimists would have anticipated Hill and Hillova found that only the chick fibroblasts exposed to undigested DNA from transformed cells are transformed and begin to produce virus. Moreover the virus produced is of the same antigenic subgroup as the virus used originally to transform the donor rodent cells and the virus produced by cells exposed to temperature sensitive SR.RSV is temperature sensitive. It seems clear therefore that transformed cells do indeed contain at least one DNA provirus which can be isolated in an infectious state.

With so many research groups now working with the RNA cancer viruses it should not be long before these data are independently confirmed.

killing activity reappears. The manner in which the colicin encompasses the fission of the 16S RNA is still unknown. The dependence of the reaction on the presence of 50S subunits has also now been reported by Bowman (*FEBS Lett.*, **22**, 73; 1972).

NEUROENDOCRINOLOGY

Neurones and Hormones

from a Correspondent

THE nature and significance of the connexions of the hypothalamus, the sites of action and effect of gonadal steroid hormones on the neural structures which control the secretion of pituitary gonadotrophins, and the significance of the high concentration of monoamines in the hypothalamus were the principal themes of the neuroendocrinology symposium held at the University of Oxford on April 6.

The symposium, part of the spring meeting of the Anatomical Society of Great Britain and Ireland, missed the wisdom and subtle wit of the late Professor G. W. Harris by whom it was conceived and planned. The elegance and precision of Harris's experiments, discussed by Dr B. T. Donovan (Institute of Psychiatry, Maudsley Hospital), substantiated the neurohumoral hypothesis of the control of the anterior pituitary gland and laid important foundations for modern endocrinology. It is ironic that Harris's insistence on excellence may have robbed him of an ultimate goal—the characterization of luteinizing hormone releasing factor—for the physiological methods he used to assay this factor were much slower than those used by the apparently successful American teams.

Although most of the seven contributions were prepared for an audience of general biological and medical scientists, the scholarly review of the neural connexions of the hypothalamus by Dr T. P. S. Powell (University of Oxford) will have pleased the specialist. Dr Powell focused attention on relatively novel data which suggest that the hypothalamus achieves connexions with the chief sensory association areas of the neocortex through multisynaptic pathways. These connexions could convey to the hypothalamus highly integrated visual stimuli, and, therefore, may prove to be an important component of the pathways which subserve the marked effect of light on the hypothalamo-hypophysial activity.

Professor B. Halasz (Semmelweis University) reviewed comprehensively the outstanding work by Hungarian anatomists on the determination of the area of the hypothalamus which secretes pituitary hormone releasing factors (hypophysiotrophic area) and

the investigation of the function of the deafferented hypothalamus. Although few new data were offered, this contribution stimulated lively discussion on the degree to which the hypothalamus is able to function independently of other areas of the brain.

The intriguing question of the role of the monoamines of the hypothalamus was reviewed by Dr K. Fuxe (Karolinska Institutet). Little information was added, but Fuxe did admit that the presence of axo-axonic synapses in the median eminence, the premise which underlies his theory that catecholamines exert an inhibitory influence on the secretion of gonadotrophins, could not be substantiated.

Three contributors discussed the effect of gonadal steroids on the brain with special reference to the control of pituitary gonadotrophin secretion. Dr W. Stumpf (University of North Carolina) led the audience speedily over a detailed atlas of the intracranial and extracranial structures which have an affinity for tritiated oestrogens as determined by autoradiography. Unfortunately, the functional significance of grains in relation to cells was taken for granted, and no evidence was offered to show that presence of grains was attributable to oestrogen rather than a metabolite. The fact that Dr P.

MacKinnon (University of Oxford) and her colleagues were able to demonstrate significant changes in the rate of protein synthesis (as determined by the incorporation of ^{35}S -methionine) in the manipulation of the concentration of preoptic area and median eminence during the oestrous cycle and following gonadal steroids in blood suggests that this may prove a more meaningful technique for detecting the site of action of steroids than the determination of their distribution in brain tissue.

The finding by Professor B. A. Cross (University of Bristol) and his colleagues that oestrogen increases the firing rate of certain neurones in the hypothalamus of the intact brain and in hypothalamic islands (which included the preoptic area) is especially significant in the context of the current concept that oestrogen triggers the pre-ovulatory surge of luteinizing hormone, and provides further evidence for the autonomy of the hypothalamus, emphasized previously by Halasz, in controlling the secretion of pituitary gonadotrophins. But whether the hypothalamic neurones involved in the control of gonadotrophin secretion are driven by neural influences arising within the hypothalamus, or by changes in the concentration of gonadal steroids in plasma, remains unanswered.

Action of the Cholesterol Molecule

CHOLESTEROL is known to exert a profound effect on the physical properties of membranes, both artificial and natural. When introduced into phospholipid bilayers it decreases the degree of fluidity of the interior, and it has been suggested that it does so by forming a stoichiometric complex with the phospholipids at a mole ratio of 1:1. Engelman and Rothman offer an alternative explanation of the observed effects in next Wednesday's *Nature New Biology*.

A model of the cholesterol molecule shows that the cyclic part is about twice as large as the linear hydrocarbon tail. Where the two join there is a sharp change in cross-section. Therefore the hydrocarbon chain of a lipid packed in a bilayer next to the cholesterol will have much less freedom of motion in the part lying next to the ring than that near the tail, given that all chains are known to lie essentially perpendicular to the plane of the bilayer. X-rays show that the total area of a lecithin: cholesterol pair in a mixed bilayer of composition 1:1 is close to the calculated minimum, for an all-*trans* hydrocarbon chain.

From molecular dimensions it follows that the first seven methylenes of the phospholipid in the all-*trans* state will stretch the length of the cholesterol ring. Beyond this the space available to the

hydrocarbon chain becomes much greater, and Engelman and Rothman in fact calculate that the effective cross-sectional area here will be larger by 50 per cent. Such a hydrocarbon chain will therefore have increased configurational degeneracy, which will be reduced in a neighbour not in contact with cholesterol, and reduced still more in the next molecule along and so on. This means that phospholipid chains adjacent to cholesterol are spatially largely independent of the other chains in bulk phospholipid regions, which explains why they are not expected to be involved in the cooperative changes of state (phase changes) with temperature, to which close-packed phospholipid chains are subject, and also why no phase transition is observed in a 1:1 complex, where all phospholipid chains are flanked by cholesterol.

This scheme of interaction sets no restrictions on the length of the hydrocarbon chain: everything beyond the first seven methylenes will be relatively fluid. The cholesterol thus reduces all neighbouring phospholipid chains to structural equivalence, and Engelman and Rothman suggest that it is for this reason that lipids can be exchanged between plasma membranes and the surrounding fluids, without disturbance of the basic structure.

CHEMICAL SOCIETY

Merged Chemists Meet in Manchester

from a Correspondent

THE annual meeting of the Chemical Society—now incorporating the memberships of the Chemical Society, the Royal Institute of Chemistry, the Faraday Society and the Society for Analytical Chemistry—which was held in Manchester from April 10 to 14, was the largest of its kind ever held in Britain. The ten separate symposia also represented the largest scientific programme ever presented by the old constituent societies.

In the symposium on oxidation chemistry Dr K. U. Ingold (National Research Council of Canada) discussed peroxy radicals and the autoxidation of organic compounds, and Dr D. Clark (ICI Petrochemicals Ltd) showed that in the reduction of oxygen with Cu(I) complexes the reaction proceeds through hydrogen peroxide as an intermediate. The number of excellent lectures by inorganic chemists at this symposium reflected the way in which inorganic research is being extended into organic chemistry and biochemistry. Thus, Professor J. H. Wang (Yale University) discussed the molecular mechanism for coupling oxidation to phosphorylation, and Dr R. J. P. Williams (University of Oxford) reviewed the role of transition metal ions in biological oxidations. Dr Williams pointed out that biological oxidations often require enzymes containing iron, copper, or molybdenum cations, and the two major oxidizing agents are oxygen and hydrogen peroxide. Professor G. N. Schrauzer (University of California) illuminated his audience with a description of the chemical evolution of a nitrogenase model. Model systems consisting of molybdate and a variety of sulphur ligands have been found to duplicate nitrogenase reactions to a remarkable extent.

The Dalton division symposium, devoted to new synthetic methods in inorganic chemistry, provided some of the highlights of the whole meeting. The lectures by Drs P. S. Skell (Pennsylvania State University) and P. L. Timms (University of Bristol) provided a tantalizing glimpse of a new type of reaction, that was brought about by using transition metal atoms as chemical reagents. Dr Skell showed that the chemistry of the transition metal atoms is not predictable from the knowledge of the reaction of the metals; for instance, platinum metal is unreactive but platinum atoms are highly reactive. Non-transition metal chemistry received a welcome large proportion of this symposium with lectures from Dr R. J. Gillespie (McMaster University) on new cationic forms of elements such as I, S,

Se, Te, and Hg obtained by reaction with AsF_5 or SbF_5 ; Dr M. Schmidt (University of Würzburg) illustrated procedures for chemically controlled syntheses of thermodynamically unstable new homoatomic compounds such as S_8 and S_{12} ; and Dr R. Thompson (Borax Consolidated, Chessington) reviewed high temperature synthesis of reduced boron compounds. In the radiochemistry section Dr G. W. A. Newton (University of Manchester) described his work, involving the irradiation of tungsten targets by 24 GeV protons, which has led to speculation of elements beyond $Z=105$.

The ready application of current chemical knowledge and chemical technology to human survival problems was evidenced in the synthetic and substitute foodstuffs symposium. Professor A. Spicer (Lord Rank Research Centre, High Wycombe) described the general methods used to obtain new protein sources based either on isolates from vegetable sources or by microbial protein fed on carbon sources as nutrients. This theme was further developed by Dr J. S. Gow (ICI Agricultural Division, Billingham), who outlined the production of single cell protein from a bacterial source based on methanol as feedstock. The human aspects of synthetic food were discussed by Miss D. Hollingworth (British Nutrition Foundation) who described current thought on nutritional content, texture and taste of the new protein isolates; and the legislative aspects of synthetic food were outlined by Dr M. Sharratt (Department of Health and Social Security).

The heterocyclic chemistry symposium received a good start from Dr J. Sauer (University of Regensburg) who described work involving reactions of six-membered and five-membered heterocycles such as 1,2,3,4,5-tetrazines and 1,2,4-triazines, and the cycloaddition and cycloelimination reactions of these compounds. Dr S. Gronowitz (University of Lund) reviewed the field of heteroaromatic boron compounds; and Professor L. A. Paquette (Ohio State University) lectured on the process known as uniparticate electrophilic addition and particularly on the use of chlorosulphonyl isocyanate in such additions to produce novel heterocycles such as azabullvalene.

The management for research symposium attracted a large number of participants, as did the symposium on interfaces in chemical education which, although one of the longest sessions, was probably the most faithfully attended. The number of secondary-level teachers

present in this last symposium increased by a factor of 4.5 compared with the previous year, but these eighteen teachers represented about only 1.3 per cent of all participants in the meeting. The Chemical Society as a whole, as well as the teachers concerned, would benefit if many more schoolteachers became fellows.

The education symposium began with a enthusiastic contribution on the Nuffield Combined Science and Secondary Science Projects from Mr R. J. Farr (Birley High School), who firmly advocated guided discovery rather than open discovery methods, and emphasized the importance of motivation. Mr W. C. Hall (Centre for Science Education, Chelsea College, London) and Dr J. Spice (Winchester College) discussed the Schools Council Integrated Science Project and Nuffield Physical Science respectively. A slogan for both schemes could be "education through science". Dr Spice emphasized that because an A-level pass in physical science is generally acceptable to universities in place of one in physics or chemistry, a student offering physical science can follow a wider sixth-form curriculum.

It was a pity that few tertiary-level teachers heard Professor A. K. Holliday (University of Liverpool) speak on the impact of new secondary curricula on tertiary chemical education. At present, no tertiary teacher can safely assume anything about the chemical knowledge possessed by new students. Existing A-level syllabuses overlap in the field of inorganic chemistry at one point only—the periodic table. Should full integration of practical and theory be achieved in tertiary work, and if so, how? The wider implications of chemistry must be considered; and anybody with a qualification in chemistry ought to possess skill in communication.

In the session on the schools/industry interface, Mr C. Othen (University College, Cardiff) emphasized the importance of the CIA Link Scheme and introduced the Ynysawdre Chemical Company, formed by pupils at a Rhondda comprehensive school, whose current leading product is a perfumed talcum powder. Vigorous contributions came from Dr D. Daniels (University of Bath), Mr R. P. Heppell (South Shields Grammar Technical School) and Mr A. Potts (John Deane's Grammar School, Norwich). One point to emerge was an indication that ability to communicate diminishes between the sixth form and post-graduate stages.

This expanded type of meeting has probably set the scene for the future. The large number of participants testified to its attraction. With the current interest in ecological problems the Chemical Society would do well to build upon the excellent press facilities which were in evidence at this meeting.

Pions and Heavy Ions in Radiotherapy

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This article considers the advantages and difficulties of using pions and heavy ions for cancer radiotherapy.

THE aim of radiotherapy in the treatment of cancer is destruction of the malignant growth with minimum damage to the normal tissues which are also exposed to the radiation beam. The first attempts to treat cancer by radiation were made at the end of the nineteenth century, using either a radioactive source close to the skin or a gas X-ray tube powered by an induction coil. The penetration of the radiation was poor and the treatment was not effective except for cancer in or close to the skin. Between the World Wars X-ray tubes running at 200 to 250 kV became the normal instruments of the radiotherapist, but the penetration of the beam at this energy was still not great enough for the treatment of deep-seated cancer; at a depth of 10 cm in tissue in these circumstances the dose is 20 to 45% of the dose at the skin. Consequently patients often suffered serious damage to the skin and subcutaneous tissues. This was avoided to some extent by using multiple fields through which the radiation could be directed at the tumour, but only at the expense of enlarging the volume of healthy tissue irradiated to a high dose level.

In the search for more penetrating radiation the chief emphasis until now has been placed on the development of X-ray generators at ever higher energies. A 1 MV generator was installed at St Bartholomew's Hospital in 1936, and during the 1950s and 1960s numerous betatrons and linear accelerators operating at energies from 4 to 35 MeV have been brought into use for radiotherapy. In spite of these improvements in technique, the therapist still often finds himself unable to eradicate malignant disease within the treated volume¹. And other methods of treatment are also often unsuccessful—surgery is impossible at many sites without causing excessive damage and chemotherapy has not yet fulfilled the hopes that were placed in it, and is in any case a generalized treatment which debilitates the whole body. Thus there is still scope for improvements in radiotherapy, either by improving the distribution of dose so that the tumour can be irradiated more selectively, by using a different kind of radiation which would enhance the differential response of tumour compared with normal tissue, or by altering the biological effects of existing radiations, for example, by the use of new schemes of fractionation or by combination with drugs. The third possibility will not be discussed in this article.

Dose Distributions

The ideal distribution of dose within the body is one in which radiation is absorbed only by the volume in which the tumour is thought to lie. Within this volume the dose should be nearly uniform. If beams of radiation are to be applied externally this means that the beam-edge should be as sharp

as possible, and that along the axis of the beam the absorbed dose should rise sharply at the beginning of the tumour volume and fall to zero beyond it. Fig. 1 shows the extent to which practical radiations fulfil this ideal. The depth-dose curves are normalized at a depth 10 cm, corresponding to a tumour at this position. The excessive dose to the skin incurred with 250 kV X-rays is avoided by using radiation generated at several MeV or by using γ -radiation from ^{60}Co , but the dose distribution is still far from ideal. Direct use of electrons at several MeV is valuable only for superficial sites^{2,3}. The use of heavier charged particles seems to be more favourable because they travel almost in straight lines—thus producing beams with sharp edges—and give an enhanced dose near the end of their range (the Bragg peak, with zero dose beyond). The Bragg peak is very narrow (see Fig. 1, curve 3), but can be broadened by varying the energy of the beam; with a fixed energy machine this can be done with an oscillating filter composed of a number of parallel ridges⁴. Curve 4 indicates the probable form of the depth dose distribution given by 4 GeV N^{7+} ions whose energy has been modified in this way. Without this modification curve 4 would have had the same general shape as curve 3. Curve 5 shows the measured distribution for 60 MeV π^- -mesons (negative pions).

The use of negative π^- -mesons to give the closest approach to the ideal dose distribution was first suggested by Fowler and Perkins⁵. The negative pion has a mass of $273 M_e$ (where M_e is the mass of the electron) and is slowed down in an analogous manner to the proton, giving a Bragg peak at

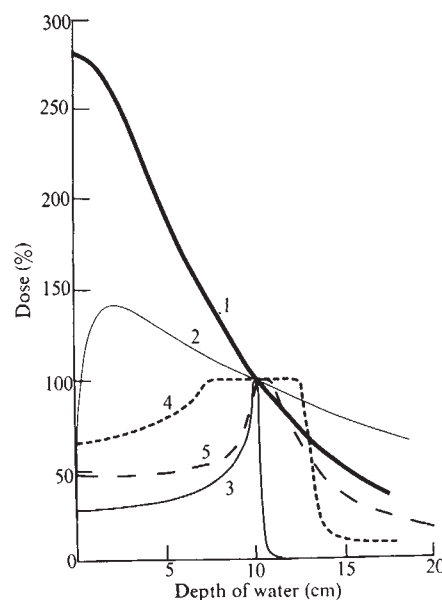


Fig. 1 Depth doses normalized at a depth of 10 cm. 1, 250 kV X-rays¹⁸; 2, 8 MV X-rays¹⁸; 3, 120 MeV protons, adapted from Larsson⁴; 4, 4 GeV N^{7+} ions with ridge filter, adapted from W. Gross and R. D. Colvett (personal communication); 5, 60 MeV π^- -mesons measured by Raju *et al.*¹⁹.

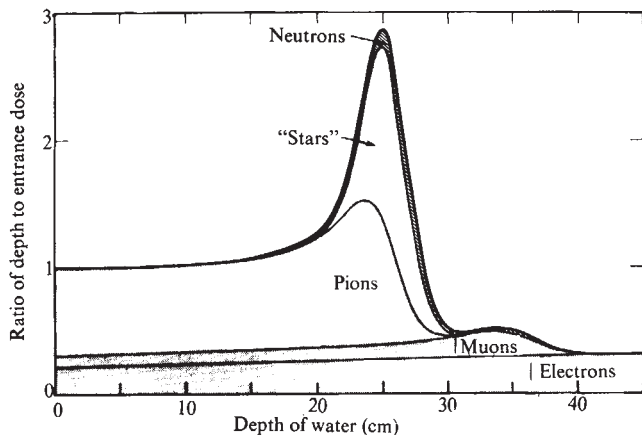


Fig. 2 Calculated depth-dose distribution for 95 MeV π^- mesons showing an analysis of the beam. $P_0 = 190 \pm 5$ MeV/c.

the end of its range. In addition to this, the pion is captured by a nucleus (usually oxygen in the case of soft tissue), once it has been brought to rest, and the resulting nuclear reaction produces several charged particles and neutrons. The average distribution of particles and energy in these nuclear reactions with oxygen, as measured by Fowler and Mayes⁶, is given in Table 1. The neutrons deposit most of their energy far

Table 1 Average Distribution of Particles and Energy in Nuclear Reactions with Oxygen

Particle emitted	Average No. of particles in each event	Mean energy per particle (MeV)	Energy deposited for each negative pion absorbed (MeV)
Proton	0.95	16	15.2
α -Particle	0.99	7.9	7.8
Heavy recoil ($Z > 3$)	0.78	5.6	4.4
Neutron	2.7	—	69

from the site of the nuclear reaction, but the charged particles have short ranges in tissue (~ 1 mm) and cause an additional increment of dose in the tumour volume. Fig. 2 shows the dose distribution calculated by Curtis and Raju⁷ for a beam of 95 MeV negative pions. The contribution from the nuclear reactions is shown in the area labelled "stars". Some of the π^- -pions decay in flight giving μ^- -mesons (muons). The contribution of these and of contaminating electrons is also shown.

Biological Properties

It would be most desirable if a form of radiation could be found to which tumour cells were more sensitive than normal cells. Unfortunately no such radiation has yet been discovered. Mammalian cells vary in their sensitivity to ionizing radiation, but only in an apparently haphazard way, with no consistent difference between normal and malignant cells. The only important difference between malignant and normal tissue that has so far been observed is that many animal tumours contain a significant fraction of hypoxic but viable cells due to the tendency of the tumour to grow beyond the limit of its blood supply⁸. Hypoxic cells are less sensitive to X-rays than aerobic cells, usually by a factor of between 2.5 and 3.5 (ref. 9). This factor is commonly known as the oxygen enhancement ratio (OER). Thus the principal radiobiological difference between normal and malignant cells is unfavourable to conventional radiotherapy. In the circumstances it is surprising that this form of treatment is ever curative. Its frequent success must be attributed to the greater recuperative power of normal tissues (which depends on the type of tumour and its tissue of origin) or to some other unknown factor such as interplay with immunity.

Heavy ions with low energies, such as those found near the

end of the range of an initially high-energy beam, or those produced by the interactions of negative pions, are often described as high linear energy transfer (LET) radiations. Linear energy transfer is usually measured by the stopping power of unit density tissue for the radiation in question. At the peak of the π^- depth-dose curve there are several particles present and hence a complex distribution of LET. Electrons, muons and pions give a low LET whereas α -particles and heavy recoils from stars give a high LET. The total calculated spectrum⁷ is shown in Fig. 3.

The biological effects of radiation with high LET are different in many respects from those of radiation with low LET, such as X-rays or γ -rays; for example, the OER falls steadily as LET rises. Another difference is the quantitative effectiveness of the radiation. As a rule a lower dose of radiation with high LET is needed to produce a given level of biological effect than is the case with X-rays or γ -rays; the ratio of doses giving identical biological effects is known as the relative biological effectiveness (RBE) of the two radiations. RBE, like OER, varies with the material irradiated and with the test of biological effect. In addition, RBE usually varies with the level of biological effort, that is, with the size of the dose delivered, whereas OER is usually independent of dose. Fig. 4 shows the way in which OER and RBE vary as a function of LET for the irradiation of aerobic cells of human origin in tissue culture, at dose levels corresponding to 250 rad of X-rays—roughly the dose used at each treatment in radiotherapy¹⁰. It is clear from a comparison of Fig. 4 with Fig. 3 that in the peak region of a beam of negative pions there will be a considerable decrease in OER and increase in RBE. The same conclusion applies to the region of the Bragg peak when heavy ions are used. Thus the use of heavy ions or negative pions will reduce the deleterious effects of any hypoxic cells existing in the tumour.

Between the skin and the tumour, however, a π^- beam has a high energy and low LET and will thus have an RBE close to unity. The same argument applies, but to a lesser extent, to high energy heavy ions. A beam of N^{7+} ions having a range of 10 cm in tissue would have a LET of about 200 MeV cm² g⁻¹ at the skin, and this would rise steadily with depth. It is doubtful, however, whether the curves of Fig. 4 would still apply to a beam of this nature. The measurements on which Fig. 4 is based were made using deuterons and helium ions of energy less than 30 MeV. Nitrogen ions of the same LET (stopping power) would have a much higher velocity and would produce a very different spectrum of δ -rays. It is known from measurements with neutrons that LET is an unsatisfactory index of radiation quality¹¹, and it seems unlikely that its use will be valid with high-energy heavy ions. The radiobiological properties of beams of heavy ions will remain uncertain until experimental observations can be made.

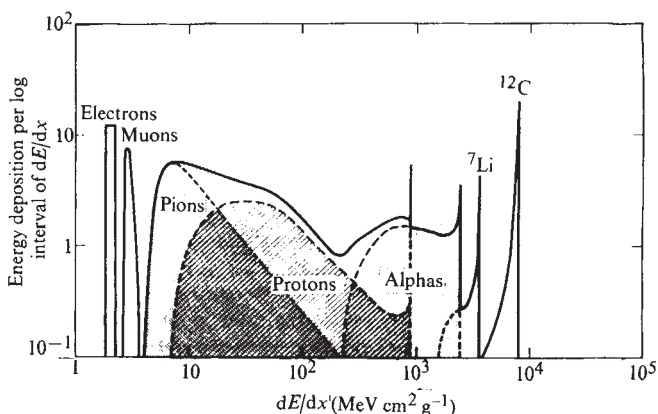


Fig. 3 Calculated LET distribution at the peak of a π^- beam. ($P_0 = 190 \pm 5$ MeV/c and depth of water = 25.5 cm.)

In both cases, however, there will be a difference in RBE between the entrance region and the peak, probably amounting to a factor of about 2 for pions and less for nitrogen ions; the effect of this will be to modify the curves of Fig. 1. If biologically effective dose were to be plotted instead of the physical quantity, absorbed dose, curves 3, 4 and 5 of Fig. 1 would be lowered in the entrance region by a factor of about 2 in relation to the 100% value at a depth. As far as is known, there is no consistent difference in RBE between normal and malignant tissues, except in so far as malignant tissues may be partially hypoxic.

There are other differences between radiations of high and low LET which may be important for radiotherapy. In general the response of cells to radiation of high LET is less dependent on modifying factors than is the case with low LET. One modifying factor, hypoxia, has been mentioned already, but there are many others which may be of equal or greater importance, for example, the position of the cell in the mitotic cycle¹², the rate of multiplication¹³, the detailed biochemical milieu and the history of previous irradiation. As far as the last point is concerned, radiation of high LET is cumulative in its effect whereas radiation of low LET loses much of its effectiveness when divided into a number of fractions spread out over several days. These differences will not be significant from a radiotherapeutic point of view unless there is a differential effect between tumour and normal tissue¹⁴. Treatment can be given up to a level at which the normal tissue close to the tumour reaches the limit of its tolerance; radiation of high LET would be better than that of low LET only if at this point it has a greater effect on the tumour. At present it cannot be said whether or not this will be the case; radiation with high LET will possibly be beneficial for some tumours growing in some sites but deleterious for others.

Experimental Work

The use of heavy ions for the radiotherapy of cancer has until now been confined to a very few centres, in particular the University of Uppsala where 187 MeV protons are used; curve 3 in Fig. 1 is derived from their measurements. High-energy protons and α -particles have been used in the United States for neurosurgery, taking advantage of the lack of scattering of the particles in tissue. There have, however, been few measurements of the biological properties of the beam in the region of the Bragg peak. Raju *et al.*¹⁵ have studied the properties of a beam of 500 MeV α -particles, using a ridge filter to spread the Bragg peak over 4 cm of tissue, and found the RBE for survival of human cells in tissue culture to be about 1.3 and the OER about 1.8, the values in the entrance region being unity and 2.5 respectively. There is therefore direct experimental evidence of a significant change in radiation quality at the peak even when the peak is spread out over several cm. The use of heavier ions such as N^{7+} would probably increase this effect because LET is proportional to Z^2 , the square of the ionic charge. Experiments to measure the radiobiological properties of 4 GeV nitrogen ions have recently been made at the particle accelerator at Princeton. The properties of still heavier ions, Ar^{18+} , for example, are at present entirely unknown; it might be that the biological effect would be constant to the end of the range, each cell whose nucleus was traversed by an ion being killed.

Comparable experiments with beams of π^- -mesons are difficult because of the very low intensities at present available. Measurements of depth dose at Berkeley and other centres have, however, confirmed the general features of the original theoretical work of Fowler⁵. Radiobiological measurements with several mammalian systems have shown that, at the peak, the OER is about 1.6 (M. R. Raju, personal communication), and the RBE greater than 2 (M. R. Raju, personal communication, and refs. 20, 21). These results

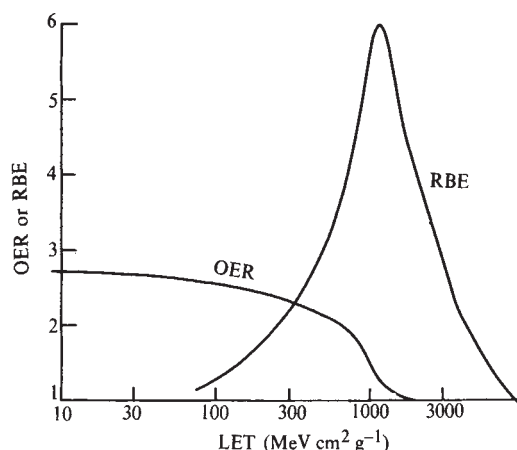


Fig. 4 RBE and OER at 50% survival (250 rad of X-rays or equivalent), from Barendsen *et al.*¹⁰.

are most encouraging and demonstrate the importance of making similar measurements at a higher dose-rate as soon as more power accelerators are brought into use.

Outlook for the Future

Any attempt to use heavy ions or negative pions for radiotherapy will be very expensive. The maximum depth in the body to which treatment needs to be given is about 15 cm, and to reach this depth with heavy ions would require about 4.3 GeV N^{7+} ions (310 MeV/a.m.u.⁻¹) or 600 MeV α -particles. On the other hand, the current required is very low, about 10^{-10} A to treat a field area of 20×20 cm to 300 rad in 5 min. Perhaps new developments in accelerator technology, such as the use of superconducting magnets, will make accelerators of this kind cheaper to build. If a machine like this were to be installed in a large therapy centre the cost for each patient could be reduced by using several extracted beams simultaneously, each being directed to a different treatment room. The angle of the beam could be fixed but different in each room, thus eliminating the need for a fully adjustable treatment head.

Pions, being a secondary radiation, require a much larger accelerated current, but the energy needed is less; pions of energy 70 MeV have a range of 15 cm of water and protons of energy about 500 MeV are required for their production; for a useful dose rate, a current of the order of 1 mA is necessary. Machines with specifications similar to this are under construction at Zürich, Los Alamos and Vancouver. In addition an advanced design is being made at Stanford for a machine to accelerate electrons to 2 GeV using a superconducting linear accelerator and the maximum possible collection efficiency for the emitted mesons.

One difficulty in the radiotherapeutic use of charged particle beams is that the depth of the peak dose in the body depends on the mass of tissue traversed; the presence of inhomogeneities (bone and air spaces) is more important than when X-rays are used. With π^- -mesons the position in the body where the particles come to rest can be monitored by measuring the emission of γ -rays and mesic X-rays or of annihilation radiation produced later by the decay of radioactive species such as ^{11}C and ^{13}N .

No dramatic increase in the overall rate of curing cancer can be expected to follow the introduction of these new forms of treatment. Patients with distant metastases are beyond the reach of radiotherapy as a curative agent. Many others are cured already by conventional radiotherapy or surgery. There remains, however, an important group of patients in whom the anatomical site of the disease precludes curative surgery, but whose tumours are resistant to radio-

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The Galapagos Triple Junction and Plate Motions in the East Pacific

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Determinations of the relative motions of the three rigid plates which meet at a triple junction lead to an estimate of the pole position for one plate if the motions of the other two are well determined. In this way, the Pacific-Nazca pole is found to a fair approximation.

SOON after the discovery of plate tectonics, McKenzie and Morgan¹ recognized that plate boundaries must eventually meet three at a time, forming what they called triple junctions. The mutual intersection of three actively spreading mid-ocean ridges is by far the most attractive type of triple junction for an initial study, but, almost by definition, such triple junctions will lie in remote corners of the oceans. We have considered the possibility that the Galapagos Rise which is known to be an actively spreading feature from 83° W to 92° W (refs. 2-4), might extend ten degrees to the west to form a triple junction with the East Pacific Rise. During a cruise of the USNS DeSteiguer (T-AGOR-12) in April and May 1970 it became apparent that there was a three-ridge triple junction at 2° 11' N, 102° 10' W, so the motion of the East Pacific Rise north of the junction is not the same as it is south of the junction. Where it is necessary to make the distinction, we will refer to the plate boundaries by the names of the two plates that they separate: the Galapagos Rise is synonymous with the Cocos-Nazca Rise; the north and south segments of the East Pacific Rise are the Pacific-Cocos Rise and the Pacific-Nazca Rise respectively.

Bathymetry

Fig. 1 shows that spreading from three rises would create a wedge shaped area of crust spread from the Galapagos Rise. Our survey (Fig. 2) shows just such a wedge, with the Galapagos spread crust about 400 m shallower and much rougher than the crust spread from the East Pacific Rise. We found that with the exception of the axial valley of the Galapagos Rise, all the features of the rises seen well away from the junction persist unchanged right up to the junction. Away from the junction the Galapagos Rise is spreading faster than most rises which develop distinct axial valleys. But if one knows (from the magnetics) the axis of the rise, one can usually find a fairly symmetrical topography, and sometimes a modest axial valley. About thirty miles from the triple junction, the Galapagos axial valley suddenly enlarges. The escarpments on the flanks of the axial valley are shallower than 1,200 fathoms and the valley floor drops to a maximum depth of 2,740 fathoms (uncorrected) at 2° 12' N, 101° 35' W (Fig. 2). We believe that this is the deepest known axial valley in the world ocean and we propose to name it Hess Deep. From its maximum development, the axial valley shoals rapidly in the last thirty miles to the triple junction. The geometrical junction, as we interpret it, falls in terrain essentially similar to the East Pacific Rise, with only broad arches to mark the three spreading rises. Dredging with a pressure-time recorder⁵ confirmed the depth

of the valley floor, and preliminary X-ray diffraction examination shows 95% of the rocks to be unaltered basalts. Bottom photographs show some pillow-like forms and abundant sessile organisms. Because the sill depth for this valley must lie about 2,000 fathoms, we conjecture that the geothermal heat flow is maintaining an adiabatic, actively exchanging water column.

There is support for the hypothesis that a similar valley has existed for all times described by similar spreading rates. The outer margin of the Galapagos spread crust is usually defined by a smooth rise followed by a sudden drop to depths greater than 2,000 fathoms (Fig. 2). We believe that these valleys may be the exported halves of the abnormally deep axial valley near the junction.

Magnetic Anomalies

Because the strikes of both legs of the East Pacific Rise are near north-south and because our survey is at low magnetic latitudes, we expect to find very little magnetic effect from normal and reversed oceanic crust. On the Galapagos Rise, however, the east-west orientation of the magnetic stripes produces anomalies of several hundred gammas at the sea surface. The magnetic bight, if one is created by the triple junction, would be expressed by the detectable Galapagos anomalies changing to undetectable East Pacific Rise anomalies along the same line that separates the Galapagos bathymetry from the East Pacific Rise bathymetry. The north-south magnetic profiles (Fig. 3) show that the magnetometer record becomes quiet at the same place that the bathymetry becomes smooth. The oldest anomaly encountered on a north-south profile through the Galapagos wedge does not establish the time at which the Galapagos Rise first became active. It had been thought⁶ that the world rise system might have been reorganized about 5 million years ago when the Galapagos Rise became active, but we would now claim that the oldest Galapagos spread material (in the Pacific) is about 30 m.y. old,

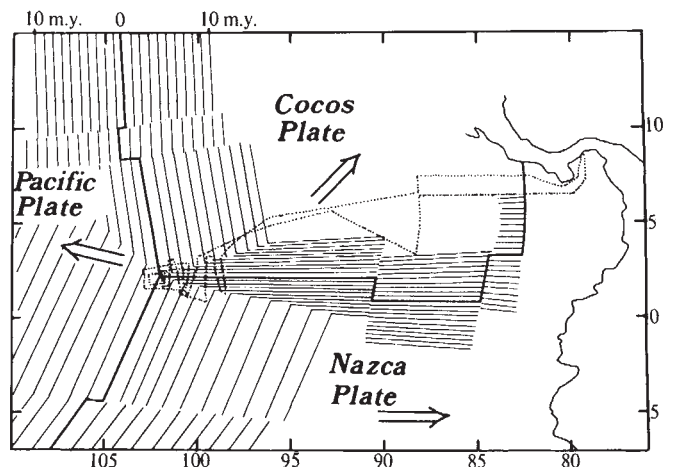


Fig. 1 Eastern equatorial Pacific plates. ---, Track of USNS DeSteiguer; —, rise axes; ---, schematic isochrons. Arrows show direction of motion of plates relative to the lower mantle¹⁴

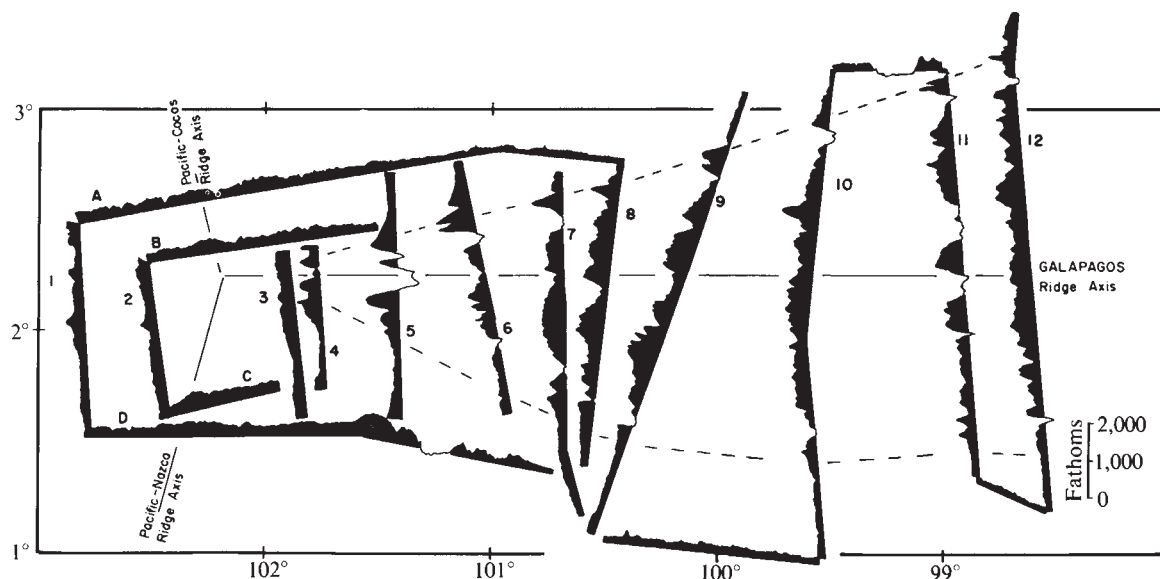


Fig. 2 Bathymetric profiles along ship track within area of triple junction survey. Base line for all profiles is 2,000 fathoms (uncorrected). —, Boundaries of crust spread from Galapagos Rise.

and that the Galapagos Rise could have been active even before 30 m.y. ago with the oldest sea floor now having disappeared down trenches.

Plate Motions

At a triple junction the vector sum of the three relative velocity vectors describing the relative motions of the three rigid plates must be zero. Thus by determining the Cocos-Nazca and Cocos-Pacific relative motions we can deduce the Pacific-Nazca pole, which cannot be directly determined at present because of lack of data. We can also deduce the Pacific-Nazca pole from the triple junction south of Easter Island and data on the Nazca-Antarctic boundaries. The Pacific-Nazca pole calculated from the two triple junctions should be the same if the plates behave rigidly.

We determined the Cocos-Nazca, Cocos-Pacific, Antarctic-Pacific and Nazca-Antarctic poles directly from spreading rates and fracture zone azimuths. We established a zone of confidence for each pole by constructing great circles perpendicular to the fracture zones, as defined by epicentres and topography, and finding the area in which they intersected. A least-squares fit to all of the available data (fracture-zone azimuths and spreading rates) was then determined with an optimization search computer program to define the pole within the zone of confidence. This program minimized $\sum_i (\text{theoretical spreading rate}_i - \text{observed spreading rate}_i)^2$ to determine either latitude or longitude, depending on the orientation of the rise, and $\sum_i (\text{theoretical azimuth}_i - \text{observed azimuth}_i)^2$ to determine the other parameter. The point at which these functions were minimized simultaneously was chosen as the best pole. (The time scale was that of Vine⁷. A paper discussing the data is in preparation.)

For the Cocos-Nazca pole, the fracture zone azimuths provide excellent latitude control, $3^\circ \text{ N} (\pm 3^\circ)$, but little longitude control. Thus the zone of confidence (Fig. 4) is a very thin, east-west band just above the equator. The longitude control, $131^\circ \text{ W} (\pm 1^\circ)$, is provided almost exclusively by the spreading rate data. So the spreading rate data and the fracture zone azimuth data may be treated independently by least squares to determine, respectively, the longitude and latitude of the pole which best fit all of the available data. This pole is located at 2° N , 131° W , and has an equatorial half-rate of spreading perpendicular to the Galapagos Rise of 4.8 cm yr^{-1} . We will use a consistent pole nomenclature here: the pole at 2° N , 131° W is the Cocos-Nazca pole, and its antipode at 2° S , 49° E is the Nazca-Cocos pole. This convention is equivalent to the right

hand rule for angular rotation vectors if the first named plate is regarded as moving with respect to the second named plate.

The Cocos-Pacific pole has previously been determined by Molnar and Sykes⁸ to be "between 20° N , 108° W and 27° N , 114° W ", and by Larson and Chase⁹ to be 40° N , 110° W with an angular velocity of $19.6 \times 10^{-7} \text{ }^\circ \text{ yr}^{-1}$. Our survey provides one additional constraint on the pole position, through the mapping of the magnetic and topographic rough-smooth boundary (Figs. 2 and 3). The slope of this boundary relative to the azimuths of the rises is equal to the ratio of the spreading rates of the Pacific-Cocos Rise and Galapagos Rise at each point on the boundary. Thus by measuring the large, easily correlated north-south anomalies and using them to determine spreading rates along the Galapagos Rise, and knowing the azimuth (078°) of the rough-smooth boundary, a spreading rate of 7.4 cm yr^{-1} on the Pacific-Cocos Rise just north of the triple junction may be solved for, even though it cannot be obtained directly because of the low magnitude latitude. Using this additional data point, the best pole was redetermined to be 39° N , 112° W , with a zone of confidence as shown in Fig. 4, with an equatorial half-rate of $11.9 \pm 3.0 \text{ cm yr}^{-1}$.

The Antarctic-Pacific pole, describing spreading on the Pacific-Antarctic Rise, was determined by Morgan¹⁰ to be at $71^\circ \text{ N} (\pm 2^\circ)$, $62^\circ \text{ W} (\pm 6^\circ)$ with an equatorial half-rate of $5.7 \pm 3 \text{ cm yr}^{-1}$. Le Pichon¹¹, using fracture zone azimuths, found an almost identical pole position of 70° N , 62° W , with an angular rate of $10.8 \times 10^{-7} \text{ }^\circ \text{ yr}^{-1}$, and, from spreading rate data, he found a pole at 68° N , 57° W , with the same angular rate. Dymek¹² re-examined the data and found a pole at $68^\circ \text{ N} (\pm 2^\circ)$, $70^\circ \text{ W} (\pm 6^\circ)$ with an equatorial half-rate of $5.5 \pm 0.5 \text{ cm yr}^{-1}$. The pole we have used (Fig. 4) is $69^\circ \text{ N} (\pm 4^\circ)$, $66^\circ \text{ W} (\pm 10^\circ)$, with an equatorial velocity of $5.6 \pm 0.5 \text{ cm yr}^{-1}$.

For the Nazca-Antarctic pole, which describes spreading on the Chile Rise, the fracture-zone azimuths give good longitude control, $84^\circ \text{ W} (\pm 7^\circ)$, but poor latitude control, $48^\circ \text{ N} (\pm 20^\circ)$ (Fig. 4). But the range in equatorial half-rates, found when the spreading rate data is included, is small, $3.1 \pm 0.1 \text{ cm yr}^{-1}$, because the range in possible pole positions is located far from the Chile Rise. When both spreading rate and fracture zone azimuth residuals are minimized, the best pole is determined to be at 49° N , 87° W , with an equatorial half-rate of 3.0 cm yr^{-1} . We note that of the four poles determined directly, the Nazca-Antarctic pole is the least well known in the sense that there are few fracture zone and spreading rate data available on the Chile Rise. The error limits given here are valid for the available data, but it is possible that further data could lead to a quite different pole position and error limits.

We now use the sum of the relative velocity vectors around the two triple junctions to find the Nazca-Pacific pole. The range in poles found from taking the vector sum around the Galapagos triple junction is $57^\circ \text{N} (\pm 25^\circ)$, $90^\circ \text{W} (\pm 40^\circ)$, with an equatorial half-rate of $8.1 \pm 2.0 \text{ cm yr}^{-1}$, with a best pole at 56°N , 94°W , and an equatorial half-rate of 8.8 cm yr^{-1} . The range found from summing around the Easter Island triple junction is $61^\circ \text{N} (\pm 5^\circ)$, $75^\circ \text{W} (\pm 10^\circ)$, with an equatorial half-rate of $8.2 \pm 0.6 \text{ cm yr}^{-1}$, and the best pole is at 62°N , 76°W , with an equatorial half-rate of 8.5 cm yr^{-1} (Fig. 4). The rough-smooth boundary does not give the same consistency of spreading rates to the south as to the north, and so a precise spreading rate could not be determined, but a possible range in half-rates of from 6.4 to 7.8 cm yr^{-1} just south of the Galapagos triple junction was found. This is compatible with both Nazca-Pacific pole positions and angular velocities.

The two Nazca-Pacific poles are the same within the limits of error arising from uncertainties in the other pole positions, and we conclude that the rigid-plate hypothesis is valid in this area. Because of the large error limits, however, the possibility of small internal plate deformations cannot be ruled out. These error limits were calculated by varying each of the cartesian angular velocity vectors through its entire range while holding the others constant. The wide range in pole positions found by summing around the Galapagos triple junction is caused by the

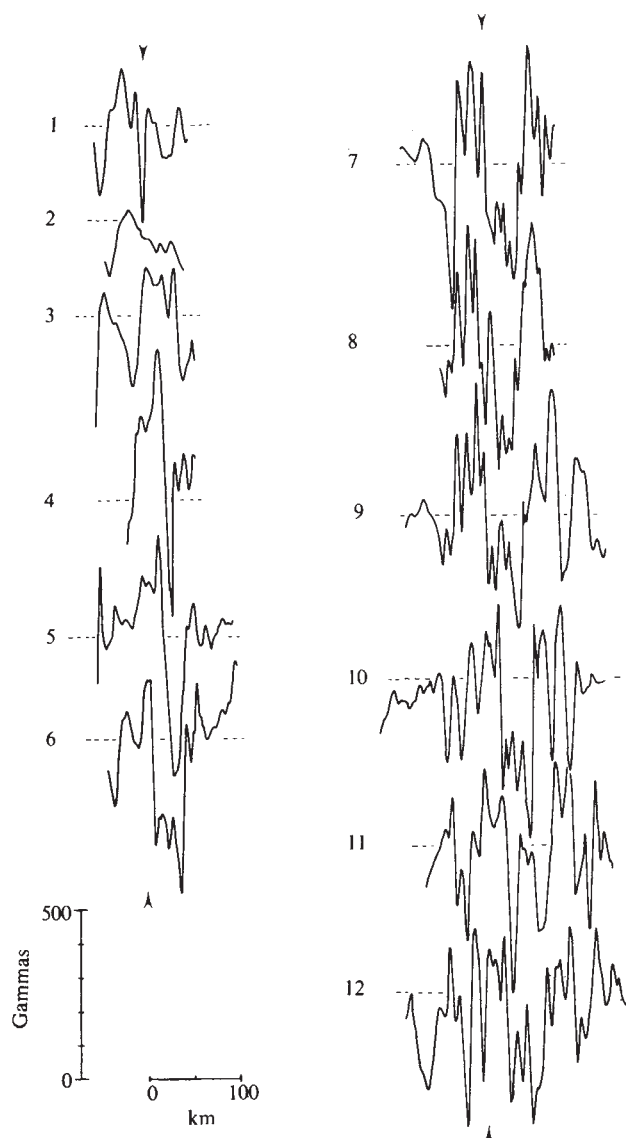


Fig. 3 Residual magnetic profiles across Galapagos Rise. Positive values are downward for easier comparison to high latitude anomalies. Profiles indexed in Fig. 2.

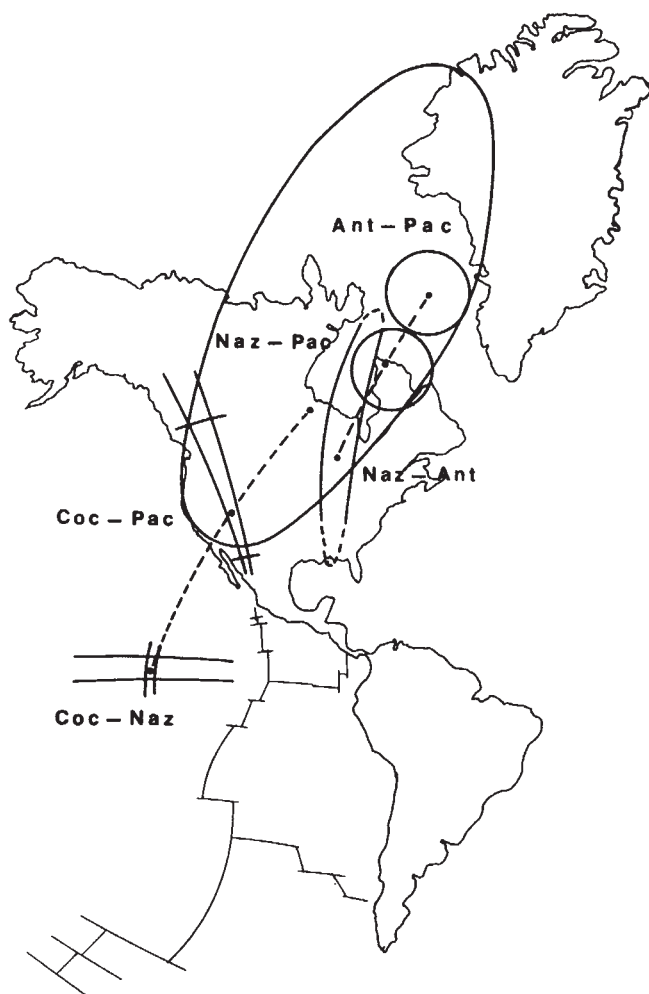


Fig. 4 Plate boundaries and relative rotation poles of the East Pacific. The ellipses represent error limits. ---, Great circles.

uncertainty in the location of the Cocos-Pacific pole, which is a result of unfavourable geometry, uncertainties in the available data and a lack of data near the equator.

We note that the apparent trend in epicentres at 4°S (ref. 13) implies that the Nazca-Pacific pole is located west of our locations. A detailed mapping of the transform faults, as well as the determination of spreading rates along the Nazca-Pacific Rise (which appears to be the fastest spreading rise in the world) would permit a direct determination of the Nazca-Pacific pole and thus provide a further test of the rigid-plate hypothesis.

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Microcrystals of a Modified Fibrinogen

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Fibrinogen forms microcrystals after cleavage with an enzyme from *Pseudomonas*. Electron microscope images reveal a close relation between these structures and fibrin. The results support the view that no major intramolecular rearrangement occurs in the fibrinogen-fibrin conversion.

FIBRINOGEN is a large molecule (molecular weight 340,000) with a complex structure; electron microscopy has revealed three globular units connected by thin rods¹. The molecule appears to be about 475 Å long by shadow casting, but this length has been reported to depend on pH². When clotting occurs, thrombin cleaves negatively charged peptides from fibrinogen, which then aggregates to form the fibrin clot. Fibrin has a striking axial repeat of 225 Å, and both "shrinkage" of the molecule^{1,3}, or a half-staggered arrangement⁴ have been proposed to account for the smaller axial period of the aggregate.

Ordered states of association of a number of simple, fully helical, fibrous proteins, such as collagen^{5,6}, tropomyosin^{7,8}, myosin rod^{9,10} and paramyosin¹¹ have revealed the length

and interactions of these molecules. In these studies the negative staining patterns of the polymorphic forms produced *in vitro* were used to determine the packing in the native assemblies. Until recently we had obtained only a limited number of ordered states of native fibrinogen^{12,13}. We have now discovered an enzymatically modified fibrinogen that forms microcrystals showing structural features of the fibrinogen molecule.

Crude *Pseudomonas* Protease

Precipitates of fibrinogen stored in the cold at low ionic strength formed unusual rod-like crystallites when certain bacteria were present¹³. We have cultured two strains of *Pseudomonas aeruginosa* from this material. One of these secretes a protease into the culture medium, whereas the other yields a protease from the ruptured cells. These proteases modify fibrinogen to produce the crystallites.

Digestion of fibrinogen by the action of these proteases affects the heaviest of the three chains comprising the molecule (Fig. 1b and c). As this band diminishes, an increasing proportion of microcrystals can be produced from the digest. Further digestion alters the second and third bands and only amorphous precipitates are obtained. Optimal crystal formation occurs when about 5–10% of the mass of the molecule is liberated as dialysable material or is found to be non-precipitable by TCA; this modified molecule will be referred to as fibrinogen Ps.

The difference in intrinsic sedimentation constants between fibrinogen Ps and native fibrinogen is about 0.5S. The mobility of these molecules on an agarose column ('Sephacrose 4B') was found to be similar, indicating that the frictional coefficients are approximately the same¹⁵. These measurements suggest that the modified molecule may be about 10–15% smaller in molecular weight than native fibrinogen. This figure is consistent with the loss of mass measured by chemical means.

Fibrinogen Ps clots when reacted with thrombin. Clot-abilities are in the range of 84–93% for the modified molecule. Covalent cross-linking by fibrin-stabilizing factor could also be effected with the fibrin Ps clot.

Electron Microscopy of Aggregates

The structure of fibrinogen and fibrinogen Ps aggregates depends critically on the ionic conditions of precipitation^{12,13}. At high ionic strength (greater than ~0.1) where charges are shielded, native fibrinogen precipitates as fibres which appear indistinguishable from fibrin (Fig. 3). Fibrinogen Ps precipitated in these conditions or clotted with thrombin has a striation pattern closely resembling that of the native protein (Fig. 3). These observations support the picture of the overall similarity in the structure of fibrinogen Ps and the native molecule.

At low ionic strength (less than ~0.05) where charges are not shielded, native fibrinogen forms fibres which appear aperiodic in the electron microscope. In contrast, fibrinogen Ps precipitates in these conditions as highly ordered rod-shaped crystallites.

The most common microcrystals observed are called "obliquely striated" (Fig. 2a). These crystallites have a blunt rod-like shape and are characterized by striations separated by about 90 Å and inclined to the crystal axis by about 17°. Frequently the striations appear "beaded". A number of images may be distinguished which show the striations related more or less prominently to a set of transverse bands repeating every 450 Å. The symmetry of the structures is p2.

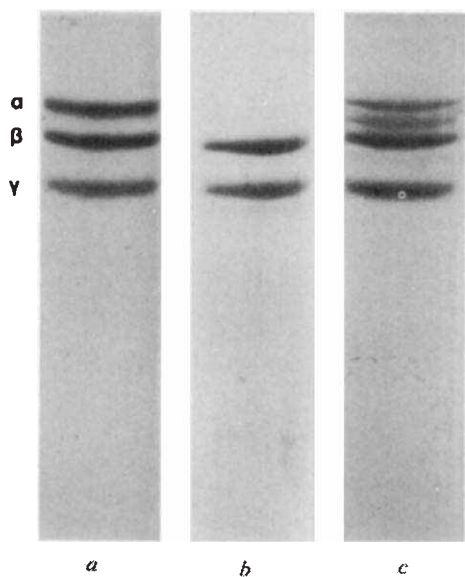


Fig. 1 SDS polyacrylamide gel electrophoresis¹⁴ patterns of fibrinogen and fibrinogen Ps reduced with mercaptoethanol. *a*, Native fibrinogen; *b*, fibrinogen digested to the point of optimum crystal formation with the protease from ruptured cells; *c*, fibrinogen digested with supernatant protease. In a typical digestion experiment, 0.5 ml. of crude enzyme solution is added to 5 ml. of a 5 mg/ml. solution of fibrinogen in 0.3 M KCl, 0.03 M phosphate, pH 7.5, and incubated at 25°C for 30 min. Phenylmethylsulphonyl fluoride is added to terminate the reaction. The reaction mixture is dialysed against 0.005 M KCl, 0.01 M MES, pH 6.5, to precipitate the protein. The course of digestion of the α chain, the most slowly moving band, is somewhat different for the two proteases. In dissociating conditions (mercaptoethanol and SDS), no high molecular weight breakdown products are found with the protease from ruptured cells. In contrast, a new band of molecular weight about 62,000, intermediate between the α and β bands, is formed with the supernatant protease.

The axial period may be taken as 900 Å (2×450 Å) for comparison with the other form shown in Fig. 2

An uncommon aggregate is the "orthogonal sheet" form. This structure is a highly symmetric, two-dimensional array with structure units oriented parallel to the crystal axis (Fig. 2*b*). This form has not been found in microcrystals prepared from fibrinogen Ps, but it has been observed in precipitates of fibrinogen contaminated with *Pseudomonas*. The structure has an axial period of 900 Å, and a lateral spacing of 90 Å, and appears to have symmetry *cmm*.

Images are often observed showing transitions from one structure to another in the same crystal. One common structure, found at the edge of the obliquely striated form, shows a simple axial repeat of 225 Å (see Fig. 2*a*). Another axial band pattern resembling that of fibrin has been observed with

the orthogonal sheet form. Various photographic superpositions of the orthogonal sheet and obliquely striated structures can account for many of the images observed. Although the detailed relation between the structures has not yet been established, an inference from the present results is that many of the polymorphic forms may be built from different packing arrangements of the same basic molecular array. It is possible that the orthogonal sheet form—or one closely related to it by tilting—may be a fundamental array which generates a number of these structures.

Relationship between Fibrinogen, Fibrinogen Ps and Fibrin

Fibrinogen Ps appears to be a proteolytically modified molecule with altered solubility and interaction properties. In

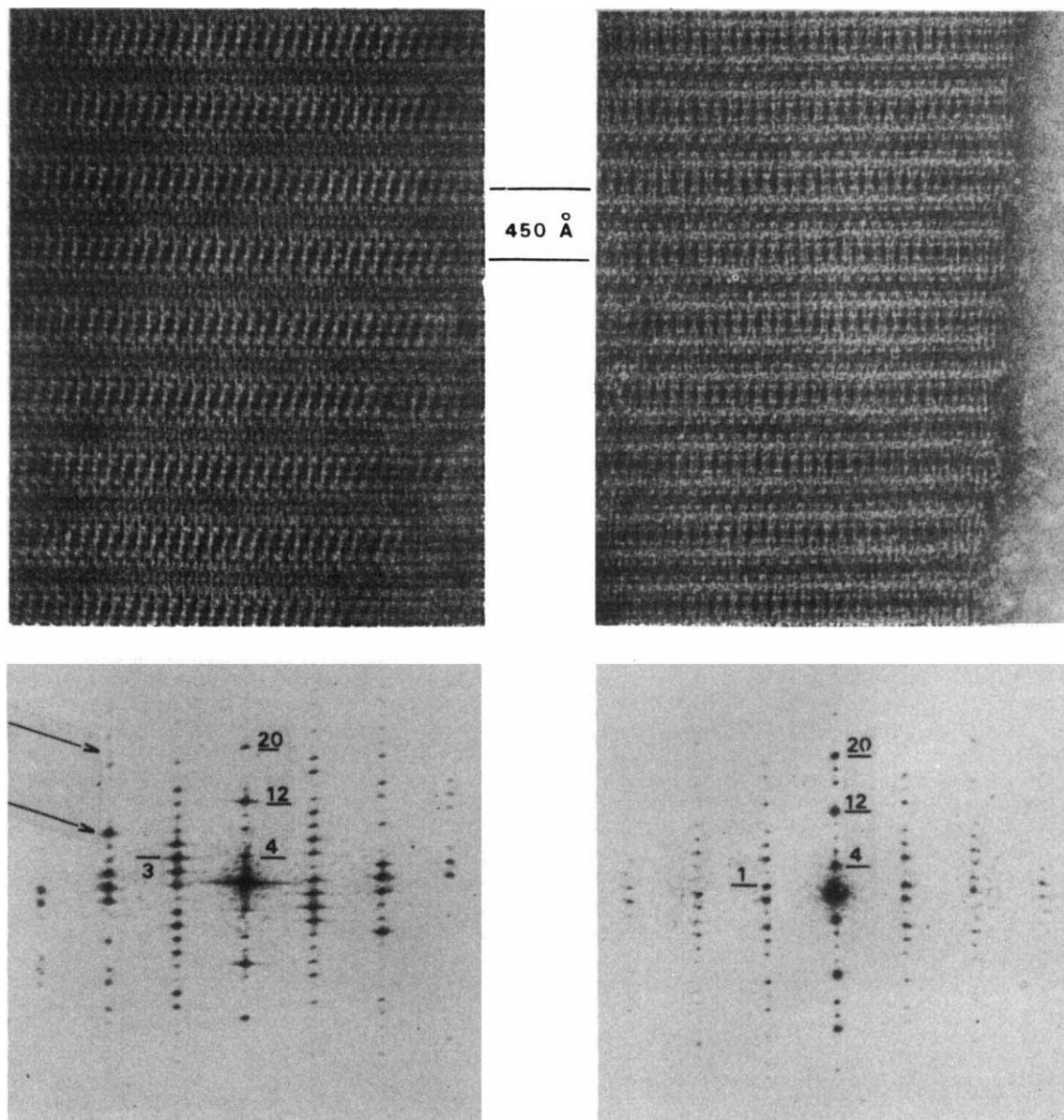


Fig. 2 Electron micrographs and optical diffraction patterns of fibrinogen microcrystals. The axial period of the structures is taken as 900 Å, and strong exposures of the optical diffraction patterns show that the order in both micrographs extends to about 25 Å. *a*, Obliquely striated form: when viewed at a glancing angle in a direction perpendicular to the axis, the structure shows transverse bands with a pseudo-period of 225 Å and a projected axial repeat of 450 Å. Correspondingly, the meridian of the optical diffraction pattern has strong reflexions which are orders of 225 Å. The striations at an angle of about 17° to the axis of the crystal give rise to the very strong 13 and 13 reflexions. The 75 Å separation of spherical units along the striations produces the strong twelfth order on the meridian. A set of fringes can be seen on the optical diffraction pattern and their direction is indicated by arrows. These fringes arise from the 75 Å periodicity along the oblique striations. *b*, Orthogonal sheet form: when viewed at a glancing angle in a direction perpendicular to the crystal axis, the structure shows a pseudo-period of 225 Å and a projected axial repeat of 450 Å. Correspondingly, the meridian of the optical diffraction pattern has strong reflexions which are orders of 225 Å. ($\times 220,000$.)

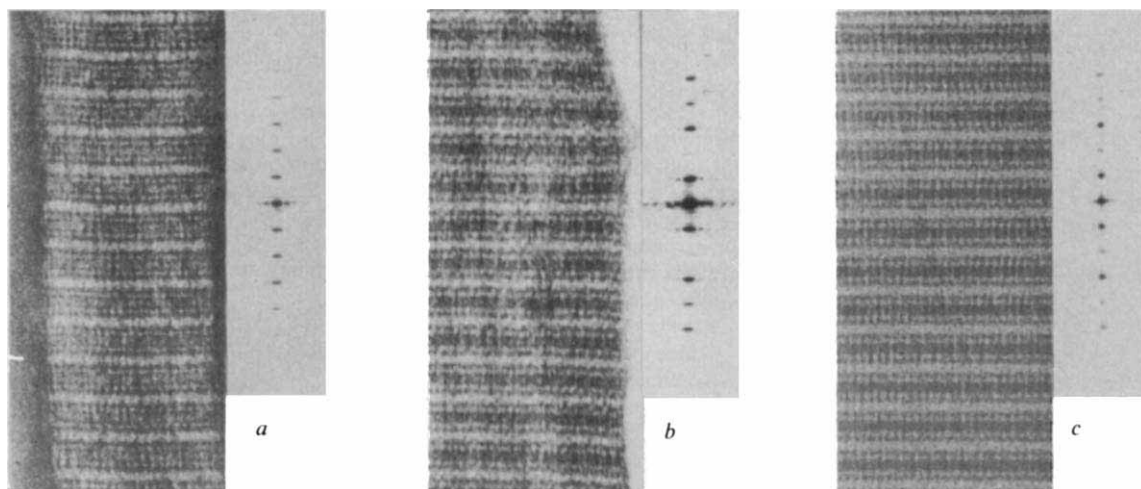


Fig. 3 Electron micrographs and optical diffraction patterns of fibrin-like forms. Native fibrinogen precipitated at high ionic strength (with 0.3 M fluoride or 0.01 M barium) appears identical to fibrin in the electron microscope. Fibrinogen Ps aggregates have the same period but a slightly different staining pattern (see text). *a*, High ionic strength precipitate of native fibrinogen; *b*, fibrin Ps (fibrinogen Ps clotted with thrombin); *c*, photographic superposition of four orthogonal sheet forms. Each is shifted 225 Å axially and 46 Å laterally to produce an image very similar to that of fibrin Ps in *b*. The effect of one superposition is to halve the axial repeat of the orthogonal sheet form causing extinctions of odd orders of 450 Å on the meridian (Fig. 2*b*). The additional superpositions reduce the lateral order ($\times 220,000$).

ionic conditions where native fibrinogen precipitates, fibrinogen Ps is more soluble. At low ionic strength where native fibrinogen forms apparently aperiodic fibres, fibrinogen Ps forms crystallites with a high degree of axial and lateral order.

Fibrinogen Ps may be as much as 15% smaller in molecular weight than native fibrinogen, but no large scale changes in structure appear to have taken place on proteolytic cleavage. If all of this mass is assumed to be lost from the α chain, as much as 40% of this chain may be missing from the altered molecule. The A fibrinopeptides are still attached, however, since fibrinogen Ps clots when reacted with thrombin. It appears that until disulphide bonds are broken—and there are twenty-eight pairs of such bridges in fibrinogen—the molecule is held intact. Support of this view comes from the similarity of the electron microscope images of native fibrin and fibrin Ps, and of the high ionic strength aggregates of these molecules (Fig. 3). Except for the possible decrease of one of the thin light staining bands, the period and negative stain distribution in the two kinds of aggregates are very similar. The small mass difference between the two molecules may account for this difference in stain exclusion.

A number of images show structure units, which are most clearly seen in the orthogonal sheet form. The distribution of mass in the projected view of this structure can be interpreted on the basis of negative staining: light regions where stain is excluded would have high protein density, and dark regions of high stain uptake would be spaces in the structure. Because of the complicated shape of fibrinogen, however, the delineation of its molecular boundaries in aggregates is not straightforward as in the case of simple rod-shaped proteins¹¹. Nevertheless, a plausible inference from both the Hall-Slayter model¹ and the solution properties of fibrinogen is that the axial repeat of 450 Å in the microcrystals may correspond to the length of the fibrinogen molecule.

A significant feature of the orthogonal sheet form is the fact that a superposition of photographic transparencies of this form shifted by 225 Å gives rise to an image very similar to the band pattern of negatively stained fibrin (Fig. 3*c*). This result implies that a specific staggering of fibrinogen arrays produces the axial repeat of 225 Å and negative staining pattern characteristic of fibrin. Fibrin occurs in nature as the unique ordered aggregate of fibrinogen produced by a specific proteolytic cleavage of the molecule. These *in vitro* studies show that fibrin may be regarded as but one of the polymorphic forms of fibrinogen. Many of these forms may be generated by different packing arrangements of the same basic molecular array. On this view, no major structural change occurs in the

conformation of the molecule in the different ordered states. This evidence supports the picture that the fibrinogen-fibrin conversion involves no large-scale molecular rearrangement in the fibrin monomer.

These fibrinogen microcrystals can be analysed by three-dimensional image reconstruction techniques¹⁶ to obtain a picture of the negatively stained molecule and the packing of its ordered states. Crystals large enough for X-ray diffraction have not yet been produced, but fibrinogen gels yield X-ray diagrams which can be used to relate the electron microscope image to the structure of the native molecule.

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LETTERS TO NATURE

PHYSICAL SCIENCES

Estimating the Firing Depth of Underground Explosions

THE usual way of determining the depth of focus of an earthquake is to measure the difference, τ , between the arrival time of the direct P wave and the arrival time of the surface reflexions pP and sP. pP and sP are the signals that travel upward from the focus as P and S waves respectively are reflected at the free surface above the focus (with conversion of some S energy to P) and then travel steeply downwards (as P waves) passing close to the focus and following a path to the receiving station similar to that of P. Thus τ depends on the difference in path length between P and the surface reflexions (and hence on the depth of focus) and on the velocity structure in the crust and upper mantle above the focus. The depth of focus can be estimated because the velocity structure of the upper mantle is known. This method is difficult to use for determining the depth of firing of underground explosions using P signals with frequencies around 1 Hz: τ is small (often 1 s or less) and the effect of the recording instrument, and of absorption in the Earth's mantle, is to smear out P and pP (sP is not generated by a pure explosion source) so that the two arrivals usually overlap. Here we describe a method of applying corrections for absorption and recording instrument effects to the P signals recorded from explosions so that P and pP can be more easily identified as separate pulses and their difference in arrival time measured. Then for explosion signals recorded at long range

$$\tau \approx 2d/v$$

where d is the depth of firing and v the velocity in the overburden.

If $F(\omega)$ and $I(\omega)$ are the effects of absorption and recording system, respectively, at angular frequency ω , $I(\omega)$ will usually be known for any given recording system and $F(\omega)$ can be written as an amplitude term $\exp(-\omega T/2Q_{AV})$ and a phase shift. T is the travel time and Q_{AV} the effective average Q (quality factor) along the path between source and receiver. Carpenter and Flinn¹ suggest that T/Q_{AV} is roughly 0.8. We have used values of T/Q_{AV} of 0.6 and 0.8. The phase shift caused by absorption^{2,3} must be included because of causality considerations.

The effects of absorption and recording system may be removed from the seismogram either by generating $F(\omega)$ for any given T/Q_{AV} and dividing the spectrum of the seismogram by $F(\omega)I(\omega)$ before making a Fourier transform into time, or by performing the computations using a spiking filter designed in the time domain. Thus if $f(t)$ is the transform of $F(\omega)I(\omega)$, the required impulse response $g(t)$ of the filter is that which, when convolved with $f(t)$, gives the best approximation (in the least squares sense) to a unit impulse or spike. This avoids division in the frequency domain which may be unstable if the denominator becomes small. Robinson⁴ discusses spike filters and has published a computer program⁵ to generate them; we used his program in the computations described here.

The effect of the spiking filter is principally to replace the high frequencies removed by absorption. Above ~ 3 Hz, however, the signal/noise ratio is usually so low that the spiking filter enhances noise rather than signal. The seismo-

gram is therefore low pass filtered to reduce the effects of high frequency noise. For the examples described here the cutoff of the low pass filter is 2.9 Hz.

Figs. 1 and 2 show the application of spike filtering to four explosion signals. Fig. 1a shows the signal recorded at the array at Yellowknife, Canada (YKA) from an explosion fired on May 21, 1968, at Bukhara, USSR. Fig. 1b is the spiked seismogram (using $T/Q_{AV}=0.6$) which shows two prominent arrivals of opposite polarity separated by 1.7 s. These are almost certainly P and pP. The geology in the vicinity of the explosion is a sedimentary basin in which an overburden velocity of 3 km s^{-1} (a reasonable round figure assumption) implies a depth of firing of 2.6 km. A 47 kton explosion was fired at a depth of 2.45 km in the vicinity of Bukhara (exact time and place unspecified) to seal an oil well⁶; it seems likely that we have analysed this explosion.

This example is clearly ideal. P and pP are well separated and can be observed on the original seismogram. This separation of P and pP can be exploited to construct a spiking filter

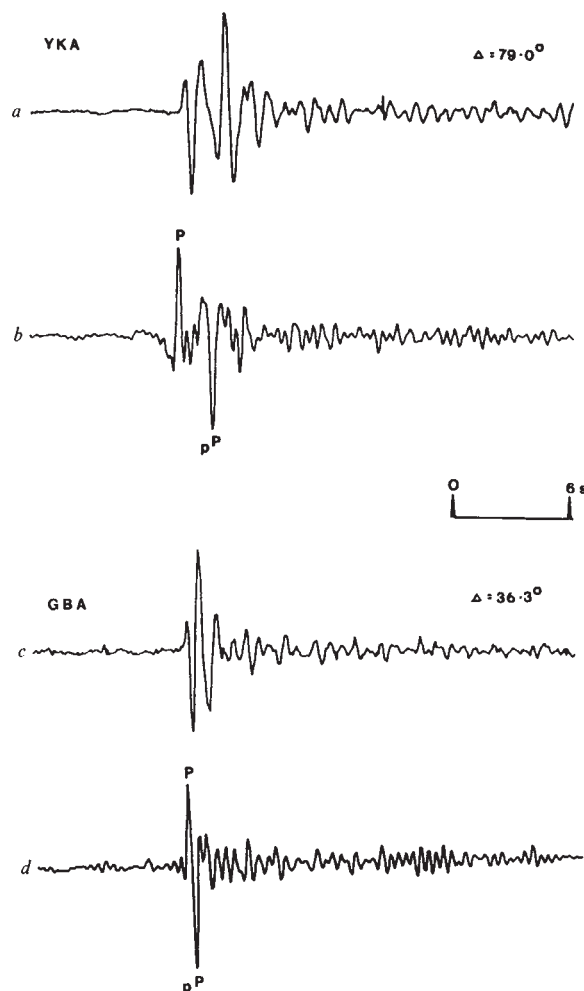


Fig. 1 Recorded and spiked seismograms for presumed explosions fired near Bukhara, USSR (a and b) and Semipalatinsk, USSR (c and d).

that takes out source effects as well as absorption and recording instrument— ϵ filter is constructed to convert the P pulse to a spike and this filter is applied to the whole seismogram; P and pP then stand out as even more prominent spikes.

The second example (Fig. 1c) is for an explosion fired on September 22, 1967, in E Kazakh and recorded at Gauribidanur, India (GBA). The spiked seismogram (using $T/Q_{AV}=0.6$) shows that the P–pP time is short (Fig. 1d) so that the two arrivals are hardly separated. But τ seems to be approximately 0.3 s. Assuming that the explosion is fired in material with the high compressional wave velocity of 5 km s^{-1} (the site is in an area which makes this value geologically reasonable), we estimate the firing depth as $\sim 0.75 \text{ km}$.

This method of estimating the P–pP time interval and hence the depths of firing of an explosion does not always work so well. Explosions in the Nevada Test Site (NTS), for example, give less convincing results principally because the source functions of most of the explosions fired at the NTS are predominantly low frequency and the average Q under Nevada seems to be so low that the high frequencies in the signal are almost completely removed.

Another complication is that many P signals from explosions are apparently not composed simply of P and pP but consist of three or more large amplitude arrivals. Fig. 2 shows the signals from the LONGSHOT (Fig. 2a) and MILROW (Fig. 2c) explosions as recorded at Eskdalemuir (EKA), Scotland. Both these explosions were fired at Amchitka Island; LONGSHOT on October 29, 1965, at a depth of 0.7 km (ref. 7), and MILROW on October 2, 1969, at a depth of 1.2 km (ref. 8). The velocity in the overburden at the LONGSHOT site has been measured as 3.38 km s^{-1} (ref. 9) so the P–pP time interval for LONGSHOT should be $\sim 0.4 \text{ s}$ and for MILROW $\sim 0.7 \text{ s}$. Arrivals at approximately these times can be seen on the spiked seismograms (using $T/Q_{AV}=0.8$) but there are later arrivals to be explained.

Our interpretation is that there are two direct ray paths between Amchitka and EKA with differences in travel time between the two paths of approximately 0.9 s; consequently, P and pP are duplicated. Two P signals (P_1 and P_2) and two pP signals (pP_1 and pP_2) can be identified on the LONGSHOT record separated by $\sim 0.9 \text{ s}$. For MILROW the time difference between the two ray paths and the P–pP time interval are almost equal so that pP_1 and P_2 nearly coincide; because these arrivals are of opposite signs they tend to cancel out. P_1 and pP_2 are thus the most obvious features of the MILROW signal, so the P–pP time interval might be taken as the P_1 – pP_2 time interval and the depth wrongly estimated as roughly twice the true MILROW depth.

This evidence of multipathing clearly implies structure in the source–receiver path, probably near the explosion source. Julian and Davies¹⁰ have shown that a descending lithospheric plate beneath Amchitka Island will produce multipathing. This may be the effect we are seeing at EKA, but we have evidence of multipathing from explosion signals recorded at EKA from the Nevada Test Site where there is no evidence of a descending plate; so a plate is apparently not the only structure that produces multipathing.

The most obvious application of the method of depth estimation described here is to the discrimination between earthquake and explosion signals. Only events that are very shallow could be explosions. The method might also be useful in spotting attempts to hide an underground test by firing a series of explosions to produce a signal that resembles an earthquake, in the way described by S. J. Lukasik in a statement to the Joint Congressional Committee on Atomic Energy, on October 27, 1971. Use of a spiking filter might reveal the individual P and pP produced by each explosion.

It is also possible to estimate the source functions of explosions if the correct value of T/Q_{AV} is used to construct the spiking filter, because the isolated P pulse is effectively the explosion source function. When pP follows very closely on P the source function can be obtained by integrating the

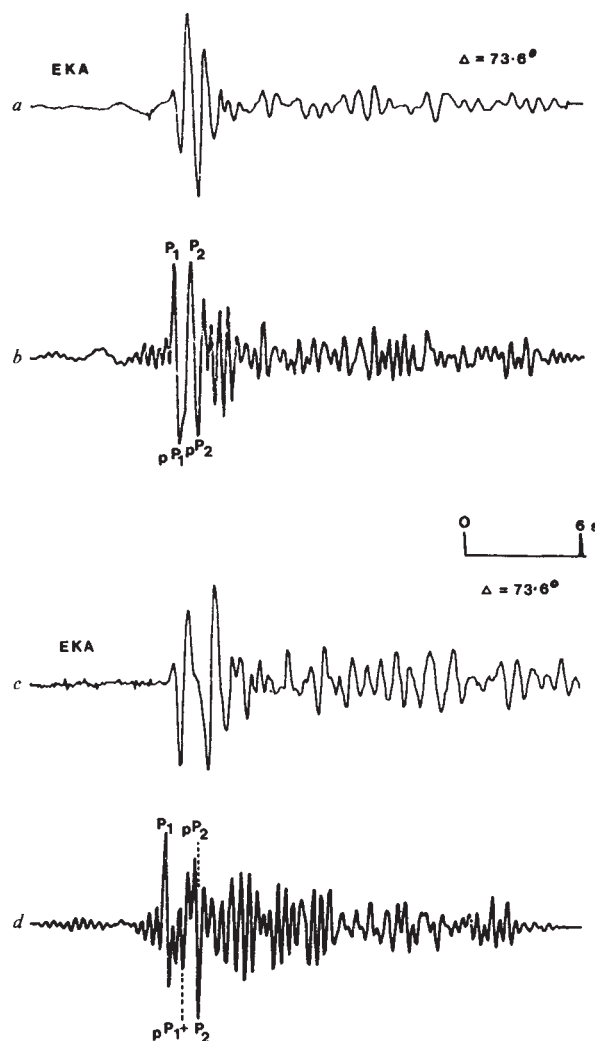


Fig. 2 Recorded and spiked seismograms for the LONGSHOT (a and b) and MILROW explosions (c and d) fired at Amchitka Island.

combined P and pP pulse. If the source function is $y(t)$ then the combined P and pP pulse after spiking is $y(t) - y(t - \tau) \approx (dy(t)/dt) \tau$ so integrating gives $y(t)$. The difficulty in estimating source functions is that T/Q_{AV} will not usually be known exactly.

The method also provides a way of getting a rough estimate of the upper limit to the yield of an explosion. Kedrovskii (see ref. 6) gives $160 W^{1/3} \text{ m}$ as the depth of burial required to completely contain an explosion of yield W . So, given an estimate of the depth of burial, the yield of the explosion that will just be contained can be deduced—although obviously the yield could be much less than this. For the E Kazakh example (Fig. 1c) the upper limit of yield is about 100 kton. Using the surface wave magnitude–yield curve of Marshall *et al.*¹¹ the yield of this explosion ($M_s = 3.3$) is estimated as 20 kton.

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Nitrogen Deficiency in the Small Magellanic Cloud

THE [NII] line at 6584 Å, which flanks H_α at 6563 Å, is often conspicuous in the emission line spectra of gaseous nebulae and galaxies. This juxtaposition allows one to obtain a ratio of line intensities (hereafter called 6584/H_α) that is free from the effects of interstellar reddening. The observations of Henize¹ and others show that among galactic planetary nebulae this ratio can range from 0 to nearly 2. On average the ionized nitrogen line is somewhat stronger in diffuse nebulae than in planetaries. Burbidge and Burbidge² have found that in the nuclei of galaxies the ratio can reach values close to 10.

Astrophysicists have generally regarded the marked fluctuations in this ratio among nebulae and galaxies as caused chiefly by excitation effects rather than by abundance differences, but Peimbert³ has shown that the spectra of M51 and M81 are best explained if the nitrogen in the nuclei of these galaxies is actually overabundant by between 2 and 6 times the solar value. We have recently made observations of the planetary nebulae in the Magellanic Clouds, obtaining a result which, like that of Peimbert, can best be interpreted as showing an anomalous nitrogen abundance. In this case, however, the evidence suggests that the Small Cloud (SMC) is deficient in nitrogen relative to the Large Cloud (LMC) or the Galaxy.

The spectra of the planetaries in the H_α region were obtained on unwidened objective-prism plates taken with the University of Michigan Curtis Schmidt telescope at the Cerro Tololo Inter-American Observatory in Chile. We exposed a Kodak 098-02 red-sensitive emulsion for 1 h behind a Schott RG1 filter. The coupled 4° and 6° objective prisms provided a dispersion of 420 Å mm⁻¹ at H_α and this, combined with the excellent seeing conditions at Tololo, successfully resolved the H_α and 6584 Å lines.

Eye estimates were made of the 6584/H_α ratio in 32 SMC and 110 LMC emission-line objects that we considered to be planetary nebulae, by the criteria that they lacked a continuum and appeared unresolved, as true planetaries must at the distance of the Magellanic Clouds (~60 kpc). Most of these have been previously classified as planetaries or probable planetaries⁴⁻⁶. A complete catalogue of the objects included in this study will be published elsewhere; here we give merely the observed percentage distribution of the 6584/H_α ratio (Table 1). For comparison we have also listed the percentage distribution adopted by Henize¹ for this ratio among a sampling of planetary nebulae in the Galaxy.

In the Small Cloud only one of thirty-two objects has a detectable 6584 Å line, and in the others the line must be very weak and may be absent. By contrast, approximately 40% of the LMC objects definitely show the nitrogen line, a proportion

comparable with that found in the galactic sample. It also appears to us that the nitrogen line is generally weak or absent in the more compact HII regions in the SMC, but this is uncertain because the 6584 Å and H_α lines become blended in diffuse nebulae at this dispersion. Slit spectra are required to validate this important point.

Table 1 Percentage Distribution of the 6584/H_α Ratio Among Planetary Nebulae in Three Galaxies

System	Number	6584/H _α		
		<0.1	0.1 to 0.5	≥0.5
SMC	32	97%	0%	3%
LMC	110	61%	15%	24%
Galaxy (from ref. 1)	49	55%	22%	22%

From these results we conclude that nitrogen must be deficient in the Small Cloud planetary nebulae. Presumably this reflects an overall deficiency of nitrogen in the stellar and interstellar components of the SMC relative to the abundance in the other two systems. The alternative explanation, that the ionized nitrogen line is not seen because of an excitation effect rather than an abundance difference, appears to require that the physical properties of the SMC planetaries differ systematically from those of the LMC planetaries. Photometric and spectroscopic studies^{7,8} have not disclosed any marked systematic differences that could account for our observations. Certainly, because the clouds are at comparable distances and the planetaries are observed to a similar limiting magnitude, we are dealing with objects having comparable luminosities, which presumably were formed at similar epochs in the past of each system. Feast⁷ has shown that if one compares the brightest planetaries in the two systems, the SMC apparently lacks the high excitation types found in the LMC, that is, those having a strong HeII line at 4686 Å. In the light of our new results, it seems that this also might represent an abundance effect.

The earlier observations of Arp⁹ and the recent photometric results of Gascoigne¹⁰ and van Genderen¹¹ have indicated that several types of stars in the Small Cloud are metal deficient compared with their counterparts in the LMC and the Galaxy. This supports our nitrogen deficiency interpretation because the production of nitrogen in stars involves the slow process of hydrogen burning of material initially enriched in carbon and oxygen¹². Therefore, the production and abundance of nitrogen is very sensitively dependent on the availability of these heavier elements in the interstellar medium and should they be underabundant an extreme deficiency of nitrogen is possible.

A deficiency of heavier elements in the SMC could be related to the rates of star formation and evolution in that galaxy. As Arp⁹ and others have noted, the Small Cloud appears to be a very young system, rich in gas and deficient in dust, in which star formation has, for some unknown reason, been retarded or perhaps commenced only recently. Consequently, the enrichment of the interstellar medium in heavier elements, through nucleosynthesis and the ejection of material from within highly evolved stars, would be far less advanced in the SMC than in the LMC or the Galaxy.

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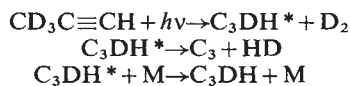
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Origin of C₃ in Comets

SEVERAL plausible parent molecules may be proposed for cometary species such as H, OH, C₂, CN and so on, but no parent molecule has been shown to produce C₃ by primary photodissociation without the intervention of secondary collisions. Swings¹ has considered the possibility that the parent molecule is diacetylene (H—C≡C—C≡C—H), and Callomon and Ramsay² have observed C₃ in absorption in the flash photolysis of diacetylene diluted with a high pressure of He. They suggest that the primary process may be C₄H₂ + *hν* → C₃H + CH followed either by decomposition of C₃H to C₃ + H or by reaction of C₃H with CH to form C₃ + CH₂. The first alternative does produce C₃ without secondary collisions, but it could not be responsible for the observations in the flash photolysis experiments in a quartz vessel because the overall process C₄H₂ → C₃ + H + CH is not energetically possible above approximately 1057 Å (ref. 3). An additional complication is that product analysis from the continuous photolysis of diacetylene has led to the suggestion⁴ of primary processes such as C₄H₂ + *hν* → C₂H₂ + C₂ and C₄H₂ + *hν* → 2 C₂H, although C₂ was not seen in absorption in the flash photolysis experiments in isothermal conditions².

Gausset *et al.*⁵ have raised the possibility of the parent molecule for C₃ being propane (C₃H₈) or propylene (C₃H₆) from which all H atoms have been removed by photodissociation. There is no evidence from laboratory work on the photolysis of either propane or propylene relating to this suggestion⁶. Here, I point out that recent evidence from several sources suggests that the related molecule propyne (CH₃C≡CH) should be given serious consideration as a potential source for C₃ in comets. Propyne is a relatively volatile compound and the simplest stable hydrocarbon containing three carbon atoms.

Recent results⁷ on the photolysis of propyne at 1236 Å show that hydrogen is formed exclusively through a molecular process. Photolysis of CD₃C≡CH yields equal amounts of D₂ and HD; the addition of inert gas reduces HD relative to D₂. I take these observations as evidence for a contribution from the following reactions



An alternate possibility is the occurrence of two simultaneous primary processes giving equal yields of hydrogen from the same carbon (D₂) as from the terminal carbons (HD). The reduction of HD relative to D₂ with increasing pressure makes this explanation unlikely because pressure is not likely to affect the relative importance of two simultaneous primary processes in photolysis with monochromatic light. Although collisional deactivation of the C₃DH intermediate occurs in the laboratory experiments, the overall process at low pressures for the non-deuterated species may be written as C₃H₄ + *hν* → C₃ + 2H₂.

I therefore suggest that a potential source of C₃ in comets is the photodissociation of propyne by solar radiation, most

probably in the region of the strong 1216 Å Lyman-α line. Although the overall process C₃H₄ → C₃ + 2H₂ is energetically possible below 1961 Å (ref. 8), a more energetic photon may be required if H₂ carries off some of the available energy.

This tentative suggestion becomes more interesting in the light of the recent discovery of propyne (methylacetylene) in interstellar space⁹. It is an attractive, although by no means established, possibility that there is a strong correlation between interstellar molecules and the stable parent cometary molecules.

Finally, I emphasize that comparable processes may occur in the photodissociation of molecules related to propyne (for example, allene, CH₂=C=CH₂) but there is no experimental evidence presently available. An attempt to detect C₃ by absorption spectroscopy in the vacuum-ultraviolet flash photolysis of propyne or allene would clarify the situation.

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Optical Polarization of X Per

It has been suggested that the variable star X Per may be the X-ray source 2ASE 0352 + 30 (refs. 1 and 2, and R. J. Brucato and J. Kristian, personal communication). Nikulin *et al.*³ report strong and variable circular polarization of Sco X-1 and HD 226868 (the possible optical counterpart of Cyg X-1), so the polarization of X Per (and its variation) is of interest. One of us (J. T., unpublished thesis) has recently developed a wide-band polarization modulator of the Dollfus type^{4,5}. A prototype using plastic retarders was used on a 30-cm Cassegrain reflector to observe the polarization of nearby bright stars, and of ζ Per and X Per. The observations on bright stars and on ζ Per show that instrumental polarization (linear or circular) is certainly less than 0.02% of the total intensity, even in the presence of several % of total intensity in polarization forms other than that being measured. Our bandwidth of approximately 1000 Å is centred between 5000 Å and 5500 Å.

We observed X Per in circular polarization on February 4 and 5, 1972. The observations are photon limited. All individual 10 min integrations yield a degree of circular polarization of less than 0.05%. If we assume the degree of circular polarization of X Per to be constant, we may average all the observations. With this assumption, and remembering the upper limit on instrumental polarization, we deduce that the degree of circular polarization of X Per is less than about 0.02%.

From sporadic observations we are not completely satisfied that the linear polarization of X Per is constant, but we cannot prove variability. The average value we obtain lies between those quoted by Behr⁶ and Hiltner⁷.

Further observations will be made with a new polarimeter when X Per emerges from the daylight sky.

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Radiation Pressure on a Test Particle in General Relativity

RADIATION pressure is believed to be the dominant force on gas in or near quasi-stellar objects (QSOs) under some conditions^{1,2}, and I have studied the acceleration of a particle to relativistic velocities by radiation pressure in an inverse-square ($1/r^2$) radiation field with the gravitational force neglected². But although arguments have been given^{3,4} that the line emission regions of QSOs are not close to Schwarzschild singularities, it is nevertheless possible that a small portion of the redshifts of QSOs may be gravitational, and it is also likely that highly ionized or stripped material lies close to the central source, where the effects of general relativity are larger. In Newtonian theory, both the radiation pressure and the gravitational force vary as r^{-2} , so that a particle of constant cross-section σ released at rest is either swallowed up or driven out, independent of its initial location. Here, I show that according to general relativity the result is different. For stationary particles the force of radiation pressure varies more rapidly than that of gravity with changing distance r to the central source. This makes stable equilibria possible. Collective effects, the effects of time variations in the strength of the radiation field and the changing opacity of the gas may well dominate the general relativistic effect discussed here. Thus, I am not proposing that the emission or absorption clouds of QSOs are necessarily stabilized in this way, but rather pointing out the presence of this stabilizing effect, in contrast with a previous suggestion in the literature⁵. Although Fowler and Hoyle⁵ do not explicitly state that stabilization could not occur in the relativistic case, they imply this by favourably comparing the stabilization in their model with that of a point source in the Newtonian approximation.

There are several discussions⁶⁻⁹ of outflowing radiation in spherically symmetric metrics. The central mass M decreases with time, the energy being carried off by radiation. In common with all the previous work, I use a radial coordinate r such that $4\pi r^2$ is the true area of a sphere of "radius" r . If u is a timelike coordinate and $M' \equiv dM/du$, then the equation $(r, \theta, \phi) = \text{constant}$ defines a stationary particle to zeroth order in M' . This definition agrees with the conventional definition of stationarity in the Schwarzschild case ($M' = 0$). Corrections to the metric because of the energy density of radiation are of the order of the fraction of mass lost in the interval r/c (ref. 9). The metric I shall use embodies these corrections, but I restrict the analysis to cases where these corrections are small in order that the definition of a "stationary" particle be unambiguous. This is almost certainly justified for QSOs, where the time scale for loss of mass by the central object through radiation

is probably $> 10^9$ yr, and the light travel time across the object can hardly exceed 10^4 yr. I use Vaidya's Radiation Schwarzschild Metric^{6,8}

$$ds^2 = -(1 - 2Mr^{-1})du^2 - 2du dr + r^2 d\Omega \quad (1)$$

where u is a timelike coordinate comparable with $(t-r)$ at large r in the Schwarzschild case. For an observer at infinity, the luminosity is $L = -M'$ (ref. 8). The energy density of radiation as measured locally by an observer with 4-velocity v^μ is⁸

$$q = (L/4\pi r^2)(\gamma + v^r)^2 \quad (2)$$

where

$$\gamma = [1 + (v^r)^2 - 2Mr^{-1}]^{1/2}; \quad v^r \equiv dr/ds \quad (3)$$

It is vital to use the exact equation (2) for q , rather than, for example, equations 10-22 of Robertson and Noonan⁹, which conserves radiation only to zeroth order in GM/rc^2 . The present results have been verified by replacing equations 10-22 of ref. 9 with one based on the accurate covariant divergence. For stationary observers ($v^r = 0$), equation 2 simplifies to

$$q = L/(4\pi r^2 |g_{00}|) \quad (4)$$

The presence of the factor $|g_{00}|$ in equation (4) is simply understood and is the cause of the enhanced radiation pressure at small r . The conservation law for electromagnetic energy may be understood as an equality in the influx and efflux of energy from any given region only when a time coordinate such as u is used, which is associated (in the limit of small M') with the invariance of the spacetime under time translations. But the local radiation density is measured by observers whose clocks are retarded (relative to u) by the factor $|g_{00}|^{1/2}$. They therefore see the flux enhanced by Hubble's "energy effect" and "number effect"^{10,11}, leading to equation (4). If k^μ is a radially directed null vector, the 4-acceleration of an isotropic scatterer of cross-section σ and mass m resulting from radiation pressure is⁹

$$\left(\frac{dv^\mu}{ds}\right)_{\text{rad}} = \frac{\sigma q}{m} \left[\frac{k^\mu}{k^\alpha v_\alpha} + v^\mu \right] \quad (5)$$

For a stationary particle, the radial component of the 4-acceleration is

$$(dr^2/ds^2)_{\text{rad}} = \sigma q m^{-1} |g_{00}|^{1/2} \quad (6)$$

If a very small term (the "induction field" of ref. 8) is neglected, Einstein's equation of motion for a particle in radial motion is, to first order in v^r ,

$$dv^r/ds = \frac{L\sigma}{4\pi r^2 |g_{00}|^{1/2} m} \left(1 - \frac{2v^r}{|g_{00}|^{1/2}} \right) - \frac{GM}{r^2} \quad (7)$$

Because $|g_{00}| \equiv 1 - 2Mr^{-1}$ decreases at small r , stable equilibria are now possible. The term in v^r supplies additional stabilization for small velocities. The factor $|g_{00}|^{1/2}$ in equation (6) appears somewhat unexpectedly as a result of the projection operators in equation (5) which are required to make $v_\alpha dv^\alpha/ds = 0$. This factor lessens the stabilization below the amount expected from equation (4) alone.

It is clear from the simple algebraic expressions in equation (7) that the place of stabilization will vary from a few to many Schwarzschild radii smoothly as the ratio $4\pi GMm/L\sigma$ varies from large values to values slightly in excess of unity. The latter case is probably more appropriate for QSOs, because rapid infall of matter is not observed although rapid ejection does occur. In examples where resonance line and bound-free opacity dominate², the central mass required to balance radiation pressure at infinity for QSOs can be as large as $10^{15} M_\odot$. It is more likely that the hydrogen is highly ionized and contributes chiefly electron scattering opacity, in which case the forces are in balance with a central mass smaller by several orders of magnitude.

The divergence of the radiation force at $r=2M$ must be treated with caution. Lindquist *et al.*⁸ show that the region inside $r=2M$ is unphysical. In addition, it would seem difficult to establish initial conditions that could lead to such a powerful outflow of radiation as is found at $r \geq 2M$. Therefore, the more plausible applications may be where $4\pi GMm$ exceeds $L\sigma$ by a few per cent, and stabilization occurs at five to a hundred times r_s , where $r_s = 2GM/c^2$. This could lead to a small gravitational component to the emission redshift in QSOs and could lead to absorption redshifts different by a few per cent from the emission redshift, as is sometimes observed, if the opacities of the emission and absorption regions are in the proper relation.

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these to conform to earlier determinations on samples from the five known geological sources in the region^{2,3}, so that comparisons could be made with the known data.

The results of the determinations (Table 1 and Fig. 1) show that the Ambitle Island and Gawa Island archaeological obsidian samples are indistinguishable, and are therefore from a common source. The closest correspondence of these samples to any of the known geological sources is with the obsidian from Talasea on the Willaumez Peninsula of New Britain. We concluded that Talasea obsidian was the raw material for the Ambitle and Gawa artefacts.

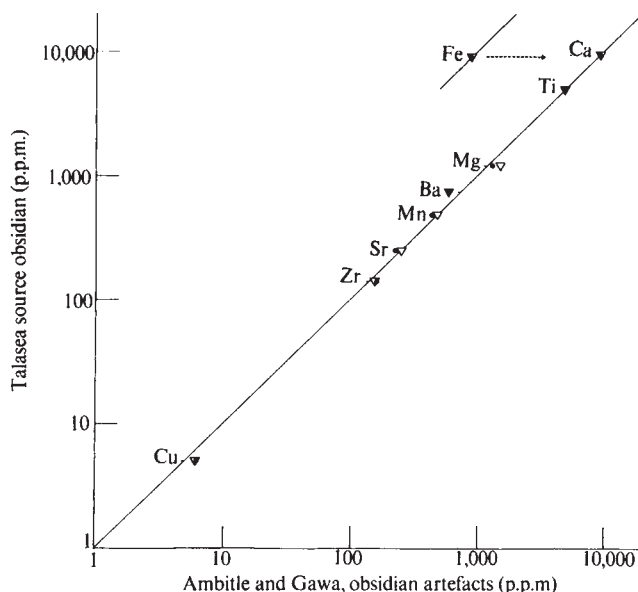


Fig. 1 The relative concentration of nine elements for the Ambitle sample (●), and the Gawa sample (▽), are practically identical. Their very close approximation to the values of elements in the Talasea source is indicated by their small horizontal distance from the diagonal, which is the line marking equal composition.

Key² demonstrated that the Talasea source provided the obsidian for the Watom Island Lapita site, which involves transport of roughly 270 km from the source. The distance from Talasea to the Ambitle site is roughly 500 km and to the Gawa site roughly 2,000 km. The ¹⁴C dates for the Gawa site are 1005 BC ± 95 (I 5747) and 825 BC ± 100 (I 5748).

Thus the data underline the effective sea-faring ability of the regions' earliest pottery manufacturers who were capable of transporting materials over long distances up to 3,000 yr ago.

The spectrographic determinations were made by Mrs M. J. Kaye of the Geology Department, Australian National University, using the procedures of Ahrens and Taylor⁴.

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First Millenium BC Transport of Obsidian from New Britain to the Solomon Islands

DURING recent archaeological work in New Guinea and the British Solomon Islands we have recovered simple flake artefacts of obsidian associated with a distinctively decorated pottery known as Lapita ware¹. This ware had a wide distribution across the south-west Pacific in the first millenium BC, and is known archaeologically from Watom Island, near Rabaul in New Britain, to Tonga, some 4,000 km to the south-west.

Table 1 Results of the Ambitle and Gawa Obsidian Determinations Compared with the Talasea Source

	Ambitle site	Gawa site	Talasea source ⁴
Sr (p.p.m.)	220	250	250
Zr (p.p.m.)	160	155	140
Ba (p.p.m.)	590	590	740
Cu (p.p.m.)	6.2	6.0	5
Mn (p.p.m.)	430	480	490
Mg (%)	0.13	0.15	0.12
Fe (%)	0.88	0.90	0.91
Ti (%)	0.15	0.15	0.15
Ca (%)	0.92	0.94	0.94

Obsidian flakes from the two sites of Ambitle, off New Ireland, and Gawa, in the main Reef Islands of the Santa Cruz group, were analysed by emission spectrography for nine elements, Sr, Zr, Ba, Cu, Mn, Mg, Fe, Ti and Ca. We chose

BIOLOGICAL SCIENCES

Warfarin and the Inhibition of Vitamin K Activity by an Oxide Metabolite

THE isolation and identification of phylloquinone oxide as a metabolite of vitamin K₁ in rats given warfarin (3- α -phenyl- β -acetyethyl-4-hydroxycoumarin) led us to investigate the possibility that the oxide is involved in the action of the vitamin or anticoagulant or both¹. We knew that phylloquinone oxide had approximately the same activity as vitamin K₁ in vitamin K deficient rats but was ineffective in those treated with warfarin². Evidence that the activity of the oxide was due to its conversion to vitamin K₁, a reaction inhibited by warfarin *in vivo*², prompted us to investigate the possibility that phylloquinone oxide, because of structural similarity, may be an inhibitor of vitamin K₁ and that warfarin exerts its anticoagulant effect by causing accumulation of the oxide.

The effect of phylloquinone oxide on the response to vitamin K₁ was tested in anticoagulant-treated animals. Male Charles River rats, 10–14 weeks old, were used. Animals given 0.4 mg sodium warfarin (Endo Laboratories) 24 h previously were injected with enough vitamin K₁ to produce a maximal rate of increase in plasma prothrombin. The effect of the oxide on the response was tested (Fig. 1a). Prothrombin was assayed according to Hjort *et al.*³ as previously used^{2,4,5}. Phylloquinone oxide was synthesized from vitamin K₁ (ref. 1). The vitamin and oxide were dissolved in 'Tween 80' and diluted with 0.9% NaCl to make solutions containing 5–12% 'Tween'; 0.1 or 0.2 ml. were injected intracardially. As little as 100 μ g of vitamin K₁ elicited a sharp increase followed thereafter by a slow increase in prothrombin concentration. Similar kinetics of response have been observed in vitamin K deficient rats by Bell and Matschner⁵ and Suttie⁶. Increasing the dose did not affect the response. As much as 1.6 mg of the oxide had little effect on the low prothrombin concentration. When 0.4 mg of oxide were injected simultaneously with 0.1 mg of vitamin K₁, normal response to the vitamin was obtained but when the dose of oxide was increased to 0.8 mg, a clear inhibition was observed. Increasing the dose of oxide to 1.6 mg did not increase the inhibition (Fig. 1a).

For inhibition to be competitive it must occur at all doses of antagonists as long as the ratio is constant. Administration of 0.2 mg of vitamin K₁ and 1.6 mg of oxide also showed a clear inhibition of response (Fig. 1a). To test higher doses, a different route of administration had to be utilized because of the limited volume that can be injected intracardially. The injection of 0.3 mg of vitamin K₁ intramuscularly produced an approximately linear response which was much slower than that resulting from intracardial administration. However, 2.4 mg of oxide clearly inhibited this response as did 3.2 mg of oxide

when administered simultaneously with 0.4 mg of vitamin K₁ (Fig. 1b).

Competitive inhibition also requires that at a constant dose of one antagonist, the inhibition can be reversed by increasing the dose of the other. With a constant dose of 0.8 mg of oxide, 0.2 mg of vitamin K₁ only slightly reversed the inhibition, but it was completely abolished by 0.3 mg of K₁. These results are consistent with the idea that phylloquinone oxide is a competitive inhibitor of vitamin K₁. However, the evidence is not conclusive because the concentrations at the active site are unknown.

The possibility that the high doses of phylloquinone oxide exerted toxic effects which inhibited prothrombin synthesis was made unlikely by injecting vitamin K deficient rats with 0.2 mg of K₁ and 1.6 mg of oxide. A normal rapid increase in plasma prothrombin occurred.

Table 1 Effect of Oxide and 'Tween' on Concentration of Vitamin K₁ by Liver

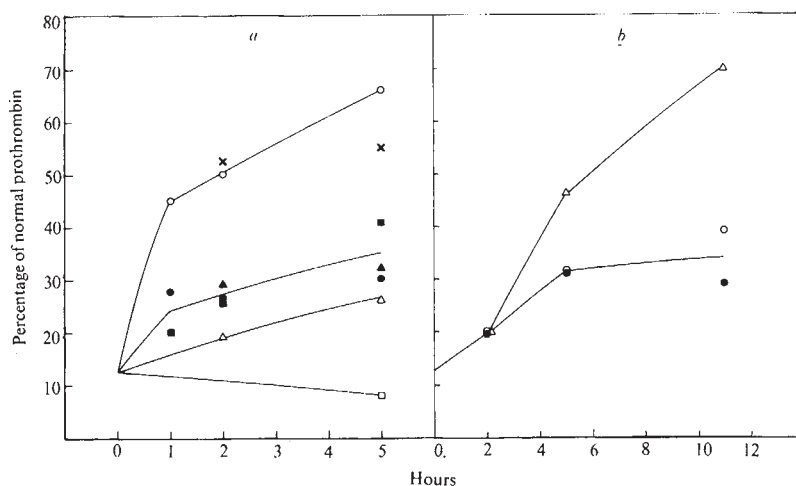
	Injected ³ H in liver at 30 min (%)
0.1 mg of ³ H-K ₁ in 12% 'Tween'	18 $\pm$ 4
0.1 mg of ³ H-K ₁ + 0.8 mg of oxide in 12% 'Tween'	30 $\pm$ 5
0.1 mg of ³ H-K ₁ in 2% 'Tween'	46 $\pm$ 10

Rats injected intraperitoneally with 0.4 mg of sodium warfarin 24 h previously were injected intracardially as shown above. 30 min after injection, the animals were killed and a portion of the liver assayed for radioactivity. Values are the averages for three animals $\pm$ s.e.

Another possibility was that high doses of phylloquinone oxide inhibited the entry of vitamin K₁ into the liver, thus blocking its effect. The oxide did not inhibit the accumulation of ³H-K₁ in the liver 0.5 h after injection (Table 1). Tritiated vitamin K₁ was prepared from tritiated menadione⁷. Liver samples were digested in NCS reagent (Nuclear-Chicago), dissolved in organic scintillator and counted in a liquid scintillation spectrometer. 'Tween 80', the emulsifying agent used, does inhibit the entry of vitamin K₁ into the liver. The injection solutions were 12% in 'Tween' in order to solubilize the high concentration of oxide. If the 'Tween' concentration was reduced to 2% (sufficient to solubilize 0.1 mg of K₁/0.1 ml.), the liver accumulation of ³H-K₁ was more than doubled (Table 1). But the stimulation of prothrombin activity in rats treated with warfarin was the same whether 0.1 mg of vitamin K was injected in a 2% or 12% 'Tween' solution. It seems that the inhibitory effect of the oxide occurs within the liver.

On the basis of these results we propose an alternative to the generally accepted hypothesis that warfarin inhibits the activity of vitamin K₁ by competing with it for an unknown active site. We suggest that warfarin exerts its effect by inhibiting the conversion of phylloquinone oxide to vitamin K₁². This

Fig. 1 Inhibition of response to vitamin K₁ by phylloquinone oxide. *a*, Rats injected intraperitoneally with 0.4 mg of sodium warfarin 24 h previously were injected intracardially with: $\circ$, 0.1 mg K₁; $\times$, 0.1 mg K₁ + 0.4 mg oxide; Δ , 1.6 mg oxide; $\bullet$, 0.1 mg K₁ + 0.8 mg oxide; $\blacktriangle$, 0.1 mg K₁ + 1.6 mg oxide; $\square$, 0.2 mg K₁ + 1.6 mg oxide; $\blacksquare$, 0.1 ml. of 5% 'Tween' solution. Each point is the average for three or more animals. *b*, Rats injected intraperitoneally with 0.4 mg of sodium warfarin 24 h previously were injected intramuscularly with: Δ , 0.3 mg K₁; $\bullet$, 0.3 mg K₁ + 2.4 mg oxide; $\circ$, 0.4 mg K₁ + 3.2 mg oxide. Each point is the average for three or more animals.



results in an increased ratio of oxide to vitamin K₁ and the oxide inhibits the activity of the vitamin by competing with it for an active site (Fig. 2). We have demonstrated that both the conversion of vitamin K₁ to the oxide and the reverse reaction occur extensively in normal rats. In animals treated with warfarin the conversion of oxide back to vitamin K₁ is blocked².

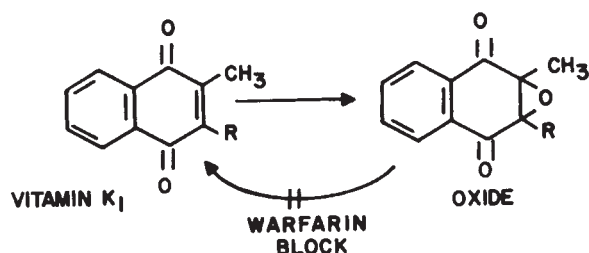


Fig. 2 Effect of warfarin on interconversion of vitamin K₁ and oxide. R, phytol side chain.

These results support the idea that vitamin K₁ and the oxide bind competitively at an active site. In contrast, there is clearly no simple competitive relationship between vitamin K₁ and coumarin anticoagulants⁸. Lowenthal *et al.*^{8,9} found that both *in vivo* and *in vitro* warfarin inhibits the stimulation of factor VII synthesis at low doses of the vitamin and anticoagulant. As the doses increase while the ratio is maintained, inhibition disappears. It was proposed that at high concentrations vitamin K₁ can enter cells by a route that is not sensitive to coumarin, but these results can also be explained by our model invoking the inhibitory effects of phylloquinone oxide. At low doses of vitamin K₁ and warfarin, a large part of the vitamin is converted to the oxide and the oxide/vitamin K₁ ratio is high. At high doses of vitamin and warfarin, a relatively smaller part of the vitamin is converted to oxide and the ratio is not high enough to cause inhibition. Preliminary evidence indicates that there are different enzymes located in different subcellular fractions catalysing the conversion of vitamin K₁ to the oxide and the reverse reaction¹⁰.

The inability of large doses of phylloquinone oxide to overcome the inhibition of prothrombin synthesis is somewhat surprising in view of its former clinical use as an antagonist of 'Dicumarol'¹¹⁻¹³. Miller *et al.*¹³ found that a commercial preparation of phylloquinone oxide (Merck and Co., Rahway, New Jersey) had little activity in dogs treated with 'Dicumarol', but large oral doses were apparently effective in patients on 'Dicumarol' therapy. The ability of the commercial preparation to overcome the inhibition of clotting factor synthesis by the anticoagulant was possibly due to contamination with vitamin K₁ since the oxide is prepared from vitamin K₁ and they are very difficult to separate¹. The phylloquinone oxide used in our studies was rigorously purified and identified¹.

The availability of humans and rats which have a hereditary resistance to warfarin has provided insight into the mechanism of action of the anticoagulant. The resistance is apparently not due to a change in the uptake, rate of excretion, or metabolism of the drug¹⁴⁻¹⁸. Hermodson *et al.*¹⁶ found that the resistant rat had twenty times the requirement for vitamin K₁ to maintain normal prothrombin levels. They postulated that the resistance results from a mutation in a receptor protein causing it to have a lowered affinity for both vitamin K₁ and warfarin. In support Thierry *et al.*¹⁷ found that ribosomes isolated from livers of normal rats contained three to five times as much labelled warfarin as those isolated from warfarin-resistant animals. The subcellular distribution of phylloquinone, however, was not significantly different in normal and resistant animals. From clinical studies on warfarin-resistant families, O'Reilly has also proposed a genetic mutation of the vitamin K-anticoagulant receptor site¹⁸. According to our proposed model for the action of warfarin, resistance in man and rats could be due to a mutation in the enzyme which converts the oxide to vitamin K₁ causing a reduced sensitivity to warfarin.

An effect of warfarin on the metabolism of vitamin K₁ does not rule out the possibility of a direct effect of the anti-coagulant on the stimulation of the synthesis of clotting proteins by the vitamin. Studies on the metabolism of vitamin K₁ and phylloquinone oxide in resistant rats should clarify this issue.

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Marker Band in One Chromosome 14 from Burkitt Lymphomas

KNOWLEDGE of the detailed pattern of fluorescence of the normal human karyotype, showing more than 200 bands per haploid chromosome set¹, has enabled us to recognize, both in biopsies and in cell cultures from several Burkitt lymphomas, an extra band in one homologue of D group chromosome pair No. 14. The deviation was seen in all analysable cells of five out of six tumour biopsies and of seven out of nine tumour cell lines examined, representing twelve different tumours from nine male and three female patients. In three tumours both biopsies and cultures were examined, and it was found that all gave results consistent in the two types of samples. Thus, two of them had the marker band in both the biopsy and culture, the third revealed the marker band absent in both cases. The remaining tumours were investigated only in biopsies or only in culture. They were positive in eight cases, negative in one. Of the twelve tumours examined altogether, ten were positive and two negative.

The appearance of the normal and the changed No. 14 is seen in Fig. 1, in which the D group shows chromosomes from two diploid cells and a tetraploid cell from three different

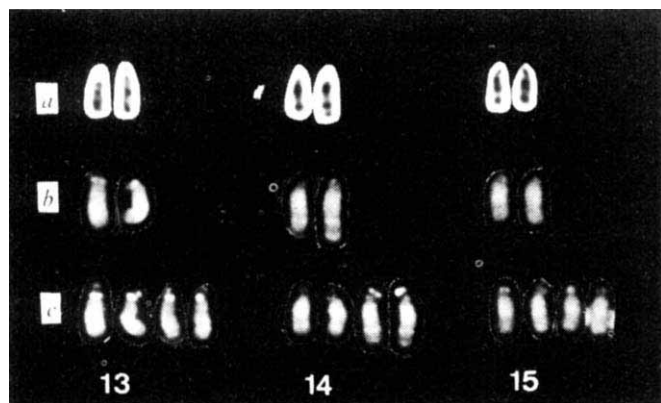


Fig. 1 Appearance of normal and changed chromosome No. 14. See text.

tumours. Fig. 1a is from a cell stained with Giemsa, after specific pretreatment giving a pattern similar to the fluorescence pattern (our unpublished results); b and c are from cells stained with quinacrine mustard (QM) using a modification of the original technique of Caspersson *et al.*². In each cell the normal chromosome is to the left and the homologue with the extra band to the right, and in Fig. 1c each homologue is represented by two chromosomes. Giemsa and QM staining shows in the changed homologue an extra band at the end of the long arm, similar to the normal fluorescing bright band near the end of the long arm (band q7¹). This extra band often appears a little less luminescent, more weakly stained with Giemsa, and somewhat narrower than the normal q7. It is impossible to decide whether the new band represents a duplication of a segment within No. 14, as an inverted duplication of the distal segment, or a translocation of a segment from another chromosome.

Table 1 Extra Marker Band at the End of the Long Arm of Chromosome No. 14 in Burkitt Lymphomas

Patient No.	Sex	Biopsies	Cultures
1	Male		+
2	Male		+
3	Male		+
4	Male		+
5	Female		+
6	Female	+	
7	Male	+	
8	Male	+	
9	Male	+	+
10	Female	+	+
11	Male		—
12	Male	—	—

+, Extra band present; (—) no extra band.

During the work we found many other markers, including the chromosome No. 10 with a secondary constriction, well known in several culture cell lines of Burkitt tumours. None of these markers, however, have been found as consistently and in so high a proportion of tumours, as the one described in No. 14. As this additional band was found in all cells in a high proportion of the Burkitt tumours examined, both in biopsies and in cultures, and as it was visible not only in QM-stained slides by fluorescence microscopy but also in Giemsa-stained slides by light microscopy, we consider it a reliable marker. The fact that a low proportion of the tumours examined did not have the marker band may mean that it had disappeared during the development of the tumour, or that there are two types of Burkitt tumours, perhaps representing different clinical entities, one with the additional band, and the other without.

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Sea Urchin Eggs Release Protease Activity at Fertilization

At fertilization, sea urchin eggs release macromolecules into the surrounding seawater. The release of these molecules (fertilization product¹) results from the lysis of the peripheral layer of cortical granules^{2,3} and possibly from other sites on the cell surface. Three macromolecules of the fertilization product have been identified: (1) a β -1,3-glucanohydrolase⁴, (2) a protein that forms part of the fertilization membrane⁵ and (3) hyalin⁶, the major constituent of an extracellular gel that surrounds the developing embryo⁷.

We now report the presence of a trypsin-like protease in the fertilization product released from fertilized or parthenogenetically-activated eggs. The activity is assayed radio-metrically (in seawater at pH 8.0) by the release of trichloroacetic acid (TCA)-soluble radioactivity from a ¹⁴C-labelled protein or colorimetrically, using the synthetic substrates α -N-benzoyl-L-arginine ethyl ester.HCl (BAEE)⁸ and *p*-toluenesulphonyl-L-arginine methyl ester (TAME)⁹. The enzymatic activity is completely inhibited by soybean trypsin inhibitor (SBTI). If SBTI is present in the seawater during fertilization the elevation of the fertilization membrane is severely impaired, suggesting that the protease may function in the elevation of the fertilization membrane.

Strongylocentrotus purpuratus gametes were shed by pouring 0.5 M KCl into opened body cavities. Egg jelly was removed by extensive washing and agitation in filtered seawater and each ml. of packed cells resuspended in 7 ml. seawater containing 0.02 M Tris (pH 8.0). Undiluted semen (25 μ l.) was added per 8 ml. egg suspension (15° C) and 5 min later the eggs were sedimented by hand centrifugation. The supernatant seawater was removed and centrifuged at 16,000g for 10 min. The clear supernatant, here designated fertilization product and containing those materials that are released by the egg and pass through the fertilization membrane, was used as the standard enzyme preparation and contained an average of 0.22 mg/ml. protein.

A radioactive protein substrate was prepared from 5 ml. (packed volume) of 20 h sea urchin blastulae incubated for 4 h at 15° C in 200 ml. seawater with 0.5 μ Ci/ml. ¹⁴C-protein hydrolysate (Schwarz). The blastulae were collected by centrifugation, extracted four times with 10% TCA (the second extraction was at 90° C for 20 min), four times in chloroform-methanol (3:1), three times in 95% ethanol and then dissolved in 20 ml. 0.05 N NaOH. The solution contained 8 mg protein/ml. (Lowry assay) and the specific activity was 224,000 d.p.m./mg. One ml. aliquots of the stock solution of radioactive protein were stored at -10° C.

For the assay, a 1 ml. portion of the radioactive substrate was thawed and the protein precipitated by adding 1 ml. 10% TCA. The precipitate was sedimented in a clinical centrifuge and dissolved in 3 ml. seawater containing 0.02 M Tris buffer,

pH 8.0. For each assay, 0.5 ml. of this substrate solution was added to 2 ml. fertilization product or 2 ml. of a solution of pancreatic trypsin (twice crystallized, Worthington) and the mixture incubated at 24° C. At 5 min intervals, a 0.5 ml. aliquot of the reaction mixture was removed and added to a tube containing 0.1 ml. bovine serum albumin (aqueous solution, 50 mg/ml.) that acted as a carrier protein during acid precipitation. The tube was mixed immediately, 0.5 ml. 10% TCA added, the tube remixed and placed on ice. The precipitate was sedimented in a clinical centrifuge and 0.8 ml. supernatant removed, placed in a scintillation vial containing 10 ml. 'Aquasol' (New England Nuclear Corp.) and the vials counted in a Beckman LS-230 liquid scintillation counter with 93% efficiency.

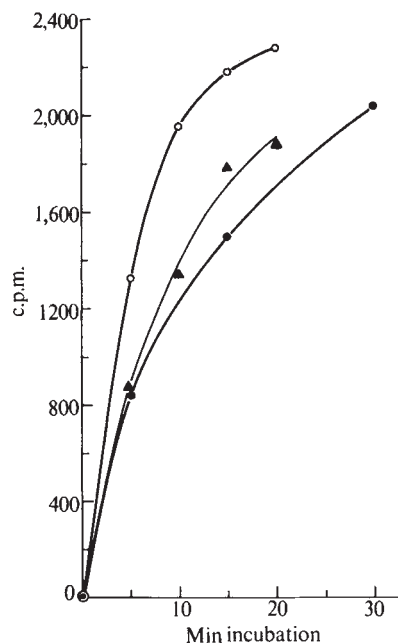


Fig. 1 Release, as a function of time, of TCA-soluble radioactivity from a ^{14}C -labelled protein by fertilization product from sperm-fertilized eggs (●), fertilization product from butyric acid-activated eggs (▲) and pancreatic trypsin (○) (0.0008 mg/ml.).

Fig. 1 shows the kinetics of release of TCA-soluble radioactivity from the radioactive protein during incubation with fertilization product or pancreatic trypsin. In both cases the rate of accumulation of TCA-soluble radioactivity decreases with increasing incubation time. Studies on the pH activity in Tris buffer over the pH range 7.0–8.5 reveal increasing activity with increasing alkalinity, the activity at pH 8 being twice that at pH 7.0. The fertilization product also hydrolyses BAEE and TAME. The activity of the standard fertilization product preparation with BAEE is equivalent to a 0.001 mg/ml. seawater solution of pancreatic trypsin (Fig. 2).

The protease activity comes from the egg and not the sperm. An equivalent amount of activity is found in the seawater surrounding eggs that were parthenogenetically activated by butyric acid (activation procedure as described by Bryan⁵ (Fig. 1)). Furthermore, in our assay conditions, protease activity cannot be demonstrated in living sperm, in living sperm agglutinated by egg jelly, or in sperm that had been frozen, thawed and homogenized in seawater.

The effects of soybean trypsin inhibitor (SBTI), limabean trypsin inhibitor (LBTI) and ovomucoid (Sigma) on the protease activity of the fertilization product was determined, using the BAEE assay. Varying concentrations of inhibitor were preincubated for 1 min with the fertilization product, the substrate added, and the rate of protease activity determined. SBTI was twenty-four times more inhibitory (on a weight

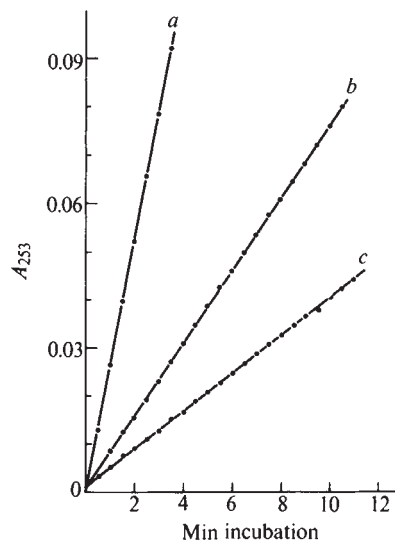


Fig. 2 Increase in absorbance accompanying hydrolysis of BAEE (Sigma) by fertilization product and pancreatic trypsin at 24° C. For the assay, 0.75 ml. of 0.0005 M BAEE in seawater (buffered with 0.02 M Tris, pH 8) was added to 0.375 ml. fertilization product or trypsin solution and the absorbance at 253 nm recorded at the indicated intervals. The stock trypsin solutions contained 0.5 or 2.5 µg trypsin/ml., equivalent to a final concentration in the assay mix of 0.16 and 0.83 µg/ml. Comparison of the fertilization product with trypsin shows the fertilization product has trypsin activity equivalent to a 1 µg/ml. seawater solution of trypsin. a, Trypsin 2.5 µg/ml.; b, fertilization product; c, trypsin 0.5 µg/ml.

basis) than the other two inhibitors. At 20 µg/ml., SBTI completely inhibited the protease activity, as measured by either the BAEE or radioactive assay.

When *S. purpuratus* or *Lytechinus pictus* eggs are fertilized in seawater containing 0.25 mg/ml. SBTI, an abnormal fertilization membrane results. The cortical granules apparently open, as the hyaline layer forms, but the vitelline layer, instead of elevating uniformly from the egg surface, remains attached to the surface at many points. The resultant fertilization membrane appears as a series of blebs, akin more to a rosette than the smooth sphere normally present. In some cases, the egg is distorted where the fertilization membrane remains attached to the surface of the egg. Ovomucoid and LBTI do not impair the formation of the fertilization membrane at 0.25 mg/ml., but do affect its formation at concentrations greater than 1 mg/ml. This lack of effectiveness of LBTI and ovomucoid compared with SBTI agrees with the weaker inhibition of the protease by these inhibitors. These observations suggest that the transformation of the vitelline layer into the fertilization membrane may depend on the proteolytic cleavage of attachments between the vitelline layer and the surface of the unfertilized egg.

The eggs of several echinoderm species parthenogenetically activate when treated with pancreatic trypsin^{10,11}. Eggs of the sand dollar *Dendraster excentricus* are very susceptible to activation by pancreatic trypsin¹¹ and pronase (D. Mazia, personal communication). These eggs, when exposed to the standard preparation of fertilization product from *S. purpuratus*, become visibly activated; the eggs elevate fertilization membranes and form hyaline layers in the same manner as sperm-fertilized eggs. If the fertilization product is preincubated for 1 min in 0.02 mg/ml. SBTI before addition of the eggs, the activation is prevented.

Neither pancreatic trypsin (0.01 mg/ml.) nor *S. purpuratus* fertilization product visibly activates *S. purpuratus* or *L. pictus* eggs. It has recently been reported that eggs of these species, when treated with pancreatic trypsin, exhibit increased amino-acid uptake and incorporation¹². However, neither of these activities increased when these eggs were treated with *S. purpuratus* fertilization product (three experiments).

The fertilization product that passes through the fertilization membrane can also be obtained from *Dendraster* eggs. It has no protease activity in the radioactive assay but does have approximately one-fifth as much BAEE activity as *S. purpuratus* fertilization product. The same activity is observed in the fertilization product obtained from *Dendraster* eggs from which the egg jelly and vitelline layer had been removed before insemination (gentle agitation of the eggs suspended in seawater containing 0.02 M dithiothreitol, pH 8.5)¹³. This low activity could result from the presence of an inhibitor. However, no inhibitory effect on BAEE hydrolysis is found when *Dendraster* fertilization product is mixed with that from *S. purpuratus* eggs. No morphological signs of activation are observed when *Dendraster* fertilization product is incubated with either *Dendraster* or *S. purpuratus* eggs.

Krischer and Chambers¹⁴ found protease activity in homogenates of *Lytechinus variegatus* eggs and showed that the activity did not change after fertilization. Using different species of sea urchins and different assays for protease activity, Lundblad¹⁵ and Mano¹⁶ observed trypsin-like activity in egg homogenates only after fertilization. To be certain that the activity found in the fertilization product does not result from cytolysis of the eggs, homogenates of unfertilized and fertilized eggs were prepared in seawater at the same protein concentration as the standard fertilization product preparation (0.22 mg/ml.). Such homogenates showed no activity in the radioactive assay.

The cellular location of the protease before fertilization is most likely in the cortical granules, which release their contents from the surface of the egg at fertilization³. Alternatively, the activity might be located on the plasma membrane and be released from it at fertilization. A TAME hydrolase has been found in plasma membrane preparations of unfertilized *Arbacia punctulata* eggs¹⁷. Finally, the protease may reside in the cytoplasm in an inactive form, be activated at fertilization, and part or all of it transported out of the egg.

Although the fertilization product of one species can activate eggs of another species, it does not seem to contain activating enzymes that can affect eggs of its own species. However, our attempts to activate with fertilization product may be unnatural, as the products are applied to the outer surface of the egg and at different concentrations than in the natural condition.

Our observations that SBTI impairs the elevation of the fertilization membrane are similar to those of Hagström¹⁸ on other species of sea urchins. The inhibition of the fertilization product protease by SBTI and the morphological effect this inhibitor has on fertilization suggests that the protease may be involved in the elevation of the vitelline layer as it transforms into the fertilization membrane.

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Are Mammalian Regulatory Systems Simple?

OHNO¹ suggests that the molecular biology of development of mammals is simpler than usually supposed. Certainly, the *Tfm* genetic variant should prove fruitful in investigating the mechanism of action of testosterone. Even accepting the validity of the proposed mode of action, however, the identification of one part of this regulatory system does not answer questions which may legitimately be asked about the molecular biology of sexual differentiation. It is not claimed that it tells us anything about the mechanism which induces the gonad to differentiate as a testis secreting testosterone in male animals but not in females; but neither does it give us any information as to how particular cells in both male and female animals come to possess the molecule (the *Tfm* protein¹) which enables them to respond to testosterone, whereas other cells do not. Again, among those cells which do produce the *Tfm* protein, there must be some prior regulation whereby the recognition of base sequences coding (in Ohno's example) for alcohol dehydrogenase and β -glucuronidase by the protein-testosterone complex (whether or not this involves Ohno's purely hypothetical homologous base sequences) occurs in kidney proximal tubule cells, but not in other target cells of testosterone where these enzymes are not induced. Yet these are the components of the regulatory system crucially responsible for differentiation between sexes and between tissues; whatever its mode of action may be, testosterone can only act as part of a switching mechanism in development by virtue of such components. In the context of classical embryology, Waddington has made a similar distinction between evocation and individuation².

In general, there is considerable genetic evidence for the interaction of many gene-action systems in controlling development. For example, the different pleiotropic effects of a gene are themselves subject to genetic control from other loci^{3,4}. Waddington's genetic assimilation experiments demonstrate that canalization of the epigenetic landscape is determined by many genes. In another developmental system, the gene *achaete* "regulates" the formation of chaetae in *Drosophila*⁵. Spickett⁶ showed the position and size of sternopleural chaetae which are formed to be determined by several interacting genes, each with a characteristic mode of action.

What, then, of the claim that regulation in mammalian development is simple? The components of the regulatory system must, by definition, be simple. If the use of a genetic variant permits isolation of one of the components of the system (thus revealing that it is under so-called single-locus control), analysis of its effects may be informative. By its very nature, however, such an investigation cannot reveal the entire sequential process of differential gene activation and repression which is responsible for differentiation, where the pattern of interactions between gene-action systems may be complex (for example, a gene may have pleiotropic effects involving different regulatory systems⁶⁻⁸). If a sufficient number of suitable

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variants could be found, the mechanisms of these interactions could be investigated^{6,9}.

Finally, avoiding polemics about genetic load and accepting the estimate of 4×10^4 genes in the human genome, the following point may still be made. If a particular gene can be involved in several different regulatory systems⁶⁻⁸, the number of interactions possible between pairs of n loci increases as $(\frac{n}{2})$. Consequently, even ignoring the currently accepted possibility of interactions occurring at several different points in the transcription-to-translation-to-effect process¹⁰, there are 8×10^8 possible gene interactions. This is sufficient to allow a considerable degree of complexity of interactions between genes, which may yet prove baffling to some investigators.

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The purpose of this study was to test this hypothesis by measuring the respiratory response to these filter conditions, and to correlate this with %COHb measured immediately after "smoking". As an index of respiratory dynamics, the breathing rate was used because it has been shown to be the most reliable and sensitive parameter⁹.

The smoking machine used, developed at the Tobacco Research Council Laboratories in Harrogate, England, has been described elsewhere^{1,2}. The rat was placed in a cylindrical restraint compartment, its nose projecting through an airtight, membranous rubber collar into a smoke chamber of 35 cm³ volume. It received a filtered cigarette smoke mixture every minute for 30 s, and fresh air for the remaining 30 s. Ten puffs were drawn from each cigarette, including the light-up puff. The four conditions presented to each of the twenty-four rats, in a Latin-square sequence, were the gas phase and the gas-vapour phase of both types of cigarettes used here. Through the transparent restraint compartment, an experienced observer was able accurately to count thoracic breathing movements during all exposures. To illustrate visually the differences in respiratory dynamics, data were collected on a separate group of rats using a photokymograph and membrane manometer. The measurement of %COHb was based on the spectrophotometric technique compiled by Richterich⁸ and adapted by one of us (A. D.). Half a minute after the last puff, the tail was punctured at about 5 mm from the tip with a disposable blood lancet. Blood (3 μ l.) was then drawn with a micropipette and mixed with a solution of 0.1 ml. 1% NH₃ in 2.5 ml. O₂-saturated water. The vessel was placed in a Beckman DB-G grating spectrophotometer and the contents balanced by 690 nm against water at 0. After the completion of haemolysis (0.5 min) and correction of the 0-point when necessary, the extinctions at 597, 577, 573, 561 and 541 nm were read, and repeated. From the extinction readings, three quotients were calculated:

$$Q1 = \frac{E(573)}{E(597)}, \quad Q2 = \frac{E(577)}{E(561)}, \quad \text{and} \quad Q3 = \frac{E(541)}{E(561)}$$

$Q1$ values served as a control against cloudiness or the transformation of Hb into other products. $Q2$ and $Q3$ represented the measurements for COHb in the total Hb. The calibration curve⁸ was compiled from forty trials with rat blood, through calculation of the $Q2$ and $Q3$ intermediate values between 0 and 100%. With this method, only 5 min of work are needed to complete the testing and calculations, only 3 μ l. of blood are necessary and the grating spectrophotometer, which can be found in most modern laboratories, makes possible the

Effect of Filtered Cigarette Smoke on Rats

THE gas phase of cigarette smoke (Cambridge and activated charcoal filter) has been shown to have a detrimental effect on several performance tests with rats, including swimming endurance¹ and avoidance response². The gas-vapour phase (Cambridge filter alone) had a significantly less toxic effect in these tests. These results were attributed to differences in respiration, in which breathing was apparently more inhibited by the irritants³ in the gas-vapour phase, resulting in a larger intake of toxic CO⁴ when the gas phase was presented alone.

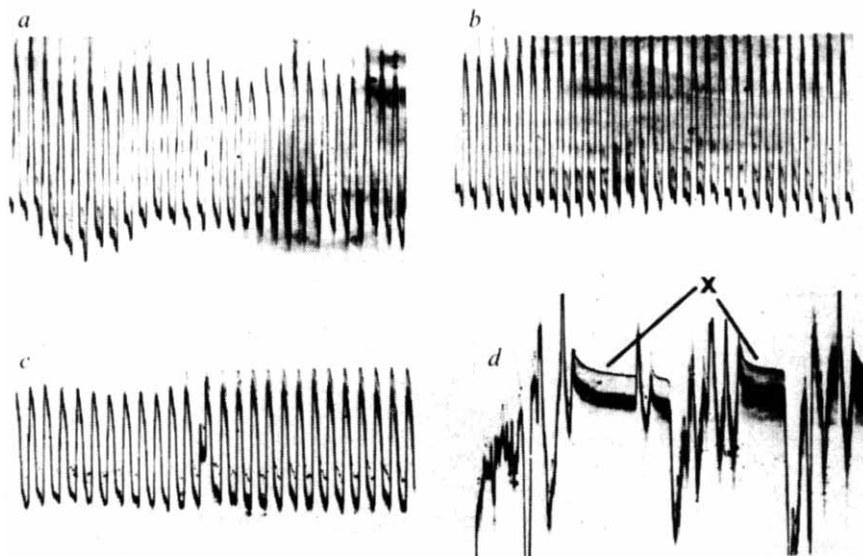


Fig. 1 Typical respiratory patterns as recorded by optical manometer: Rat Y: a, breathing in air; b, breathing in gas phase. Rat Z: c, breathing in air; d, breathing in gas-vapour phase.

measurement of more numerous and exact wavelengths. The effect of individual Hb variation among animals was minimized by each animal being its own control.

The two types of cigarettes have been described elsewhere⁷, and because they both gave similar results in regard to the gas phase vs the gas-vapour phase, they have been combined in the table presented here. The respirations during puff No. 1 were

Table 1 Average Number of Respirations in 30 s

	Average number of respirations/30 s										Avg. % COHb
	2	3	4	5	6	7	8	9	10	Total	
Gas phase	31	37	38	41	41	40	40	40	38	346	19.5
Gas-vapour	21	17	14	13	12	12	10	11	10	120	15.5

not measured, because of the act of lighting the cigarette. The separate %COHb values for all rats were submitted to an analysis of variance and statistical evaluation of the partial averages (Duncan multiple-*t* test). The differences between the gas and gas-vapour phase conditions were highly significant ($P < 0.01$) in all respects, thus confirming our hypothesis that rats are more inhibited in their breathing with the gas-vapour phase, inhaling more CO with the gas phase alone. The total respirations with the gas-vapour phase were only about one third of those with the gas phase. But the differences between the corresponding %COHb measurements, although highly significant, were not so pronounced. The answer to this could be found in analysis of the nature of the respirations, as shown in Fig. 1. The most prominent changes in the breathing pattern, caused by the gas-vapour phase (*d*), were a decrease in the total number of respirations and frequent end-inspiratory pauses ("x"). A recent study has shown a high correlation between the duration of end-inspiratory pauses and fractional increases in calculated alveolar ventilation in dogs⁸. These increases were attributed largely to an increased mixing of dead space with alveolar gas, in addition to other factors. Therefore, sporadic increases in alveolar ventilation (and increased absorption of CO through the alveoli) with the gas-vapour phase may have been responsible for reducing what might otherwise have been more substantial differences in %COHb between the two filter conditions.

Whereas the differences in respiratory response to Cambridge-filtered and Cambridge and charcoal-filtered smoke were observed here to be relatively uniform among rats, this was not the case with unfiltered or charcoal-filtered smoke, which contain the particle phase. Regarding unfiltered (full) smoke, Davis has stated that rats learn to hold their breath in the smoking machine, whereas according to Rylander, rodents "adapt" to smoking⁹. Using 15-s exposures to unfiltered and to charcoal-filtered smoke, we have found evidence to support both views. Some rats could hold their breath for the full 15 s in the course of several puffs, whereas others breathed as much as eighty-five times per cigarette. We also observed daily variations within the same animal, which has been reported for cigarette smoking/respiratory dynamics studies with humans¹⁰. The marked differences sometimes seen in respiratory response to the particle phase may be largely due to genotypic differences among individuals or groups. In an attempt to balance the respiratory differences among rats when the particle phase is presented, 5% CO₂ in the fresh air supply during smoking, or for 5 min before smoking, has been used, in order to stimulate the respiratory centre (ref. 9 and B. R. Davis, personal communication). In the behavioural research done here, where it is important to avoid possible effects from substances other than the smoke itself, we have on occasion merely prolonged the duration of the first one or two exposures. The rats often lose their breath-holding ability when forced to inhale smoke a few times at the start of a cigarette.

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Toxicity of Mercury to Phytoplankton

MERCURY in the environment has been a recent topic of discussion¹⁻⁴. According to Hammond² there are no quantitative measurements of the movement of mercury through the environment. The standards for mercury levels (5 µg l.⁻¹ for drinking water, 0.5 p.p.m. for consumable fish) which have been adopted in the United States are based on toxicity to humans but the effects of mercury on lower forms of life have received scant attention.

Clendenning and North⁵ noted a 50% reduction of photosynthesis in 4 days in the giant kelp *Macrocystis pyrifera* with 50 µg l.⁻¹ of mercury as mercuric chloride. Concentrations of 100 µg l.⁻¹ caused complete inactivation in 4 days. Boney⁶, reporting on the LD₅₀ values and the sublethal effects of mercuric chloride and n-alkyl mercuric chlorides on various species of red algae, found the toxicity of the organic compounds to increase with increasing weight of the alkyl group. The concentration of mercury salts lethal to phytoplankton was

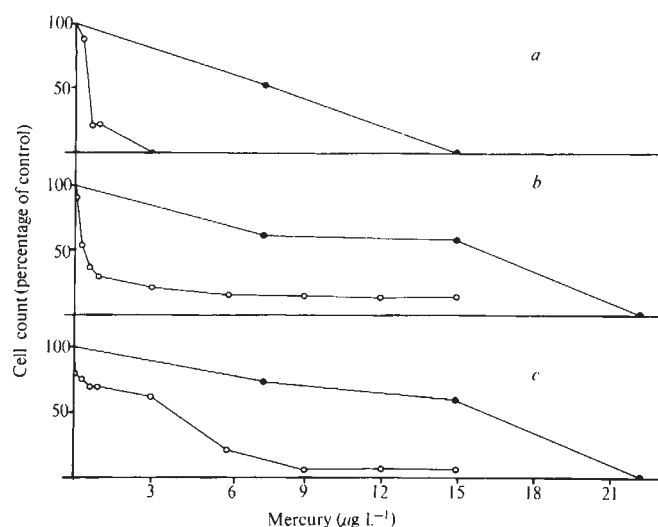


Fig. 1 The effect of mercuric chloride (●) and phenylmercuric acetate (○) on the growth of three phytoplankton species. Control growth is equal to: *a*, *Chlamydomonas* sp. 2,200,000 cells ml.⁻¹; *b*, *Chlorella* sp. 6,500,000 cells ml.⁻¹; *c*, *Phaodactylum tricornutum* 27,400,000 cells ml.⁻¹

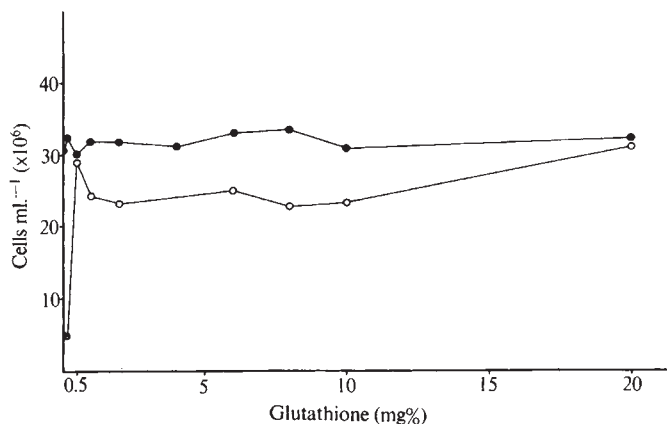


Fig. 2 Detoxification of mercury by glutathione. ●, Control; ○, mercury added at 3 µg l.⁻¹ (as phenylmercuric acetate).

Light intensity (Sylvania "cool white") was maintained at approximately 185 foot candles with a 14 h–10 h diurnal cycle. Temperature was $18^{\circ} \pm 2^{\circ}$ C. After 16 days incubation the cells were counted with a Coulter Counter Model B equipped with a 50 µm aperture tube. All the organisms were inhibited by both mercury compounds (Fig. 1). PMA was more toxic than HgCl_2 , causing inhibition at concentrations as low as $0.06 \mu\text{g Hg l.}^{-1}$ with a steep downward slope as the concentration was increased.

The addition of phenylacetate to the medium had no significant effect on the growth of *P. tricornutum*, indicating that the toxicity of PMA is due to the mercury portion of the molecule.

The inhibition of *P. tricornutum* by PMA at $3 \mu\text{g Hg l.}^{-1}$ was almost completely reversed by the addition of glutathione at 5 mg % (Fig. 2). The reversibility of mercury inhibition of thiol enzymes by the addition of monothiol compounds has been demonstrated¹³. Benesch and Benesch¹⁴ noted the

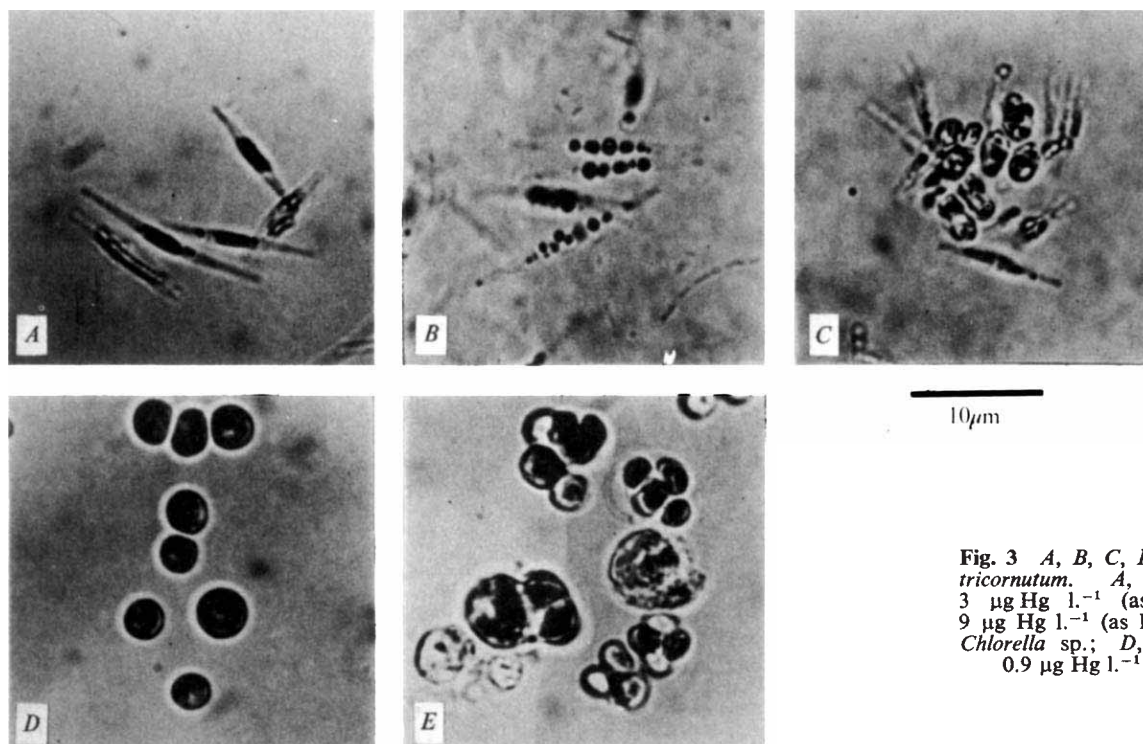


Fig. 3 A, B, C, *Phaeodactylum tricornutum*. A, Control; B, $3 \mu\text{g Hg l.}^{-1}$ (as PMA); C, $9 \mu\text{g Hg l.}^{-1}$ (as PMA). D, E, *Chlorella* sp.; D, control; E, $0.9 \mu\text{g Hg l.}^{-1}$ (as PMA).

reported to range from 0.9 – 60 mg l.^{-1} of mercury⁷. Ukeles⁸ noted that ethyl mercury phosphate (lignasan), a fungicide, was lethal to all species of phytoplankton tested at $60 \mu\text{g l.}^{-1}$ and concentrations above $0.6 \mu\text{g l.}^{-1}$ were inhibitory. Harris *et al.*⁹ found concentrations of organomercurial fungicides as low as $0.1 \mu\text{g l.}^{-1}$ effective in reducing photosynthesis and growth of natural lake phytoplankton communities and of the marine diatom *Nitzschia delicatissima*.

This laboratory has found mercury, as phenylmercuric acetate (PMA), to be inhibitory to three phytoplankton species at concentrations as low as $0.06 \mu\text{g l.}^{-1}$.

Phaeodactylum tricornutum (taxonomy discussed in ref. 10), *Chlorella* sp. (isolated from the lower Hudson River, New York¹¹) and *Chlamydomonas* sp. (isolated from Block Island Sound) were grown and tested axenically in chemically defined medium DC¹². Each was inoculated into two series of tubes containing concentrations of HgCl_2 ranging from $0.74 \mu\text{g Hg l.}^{-1}$ – $66.6 \mu\text{g Hg l.}^{-1}$ and concentrations of PMA ranging from $0.06 \mu\text{g Hg l.}^{-1}$ – $15.0 \mu\text{g Hg l.}^{-1}$. *P. tricornutum* was also tested against concentrations of phenylacetate equivalent to the phenylacetate concentration in the PMA series of tubes. All experiments were conducted in a light and temperature programmed incubator (Precision Scientific, Model 808).

formation of the mercaptide PhHgSG on the reaction of phenylmercuric compounds with glutathione. Cells grown in the presence of sublethal concentrations of mercury revealed an increased incidence of morphological abnormalities as the mercury content was increased. *P. tricornutum* became highly vacuolated (Fig. 3B) and the normally biradiate cell (Fig. 3A) assumed an ovoid shape (Fig. 3C). *Chlorella* sp. formed giant, morphologically aberrant cells (Fig. 3E).

The finding that mercury, at concentrations equivalent to those found in the sea⁴, can be inhibitory to phytoplankton demonstrates the need for a stronger research effort on the effects of sublethal concentrations of the metal on lower trophic levels. While the standards adopted for human intake of mercury may be adequate this does not dispel the possibility of much lower concentrations effecting productivity in the marine environment.

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Chronic Exposure of Rats to Methyl Mercury in Fish Protein

RECENT reports of significant concentrations of mercury in fish have emphasized the potential hazards associated with entry of this chemical as well as other heavy metals¹⁻⁸ into the food chain.

Tuna and swordfish have been more closely scrutinized than other types of fish because of their size and age, which present an opportunity for a greater build-up of mercury in their tissues than is possible in smaller, younger fish^{5,8}. In view of the current concern with mercury contamination of marine products as well as needs for satisfying world protein requirements, we report results on long-term studies in rats fed FPC (fish protein concentrate) for five generations.

The National Marine Fisheries Service prepared FPC from whole red hake (*Urophycis chuss*) by extraction with isopropyl alcohol. It was incorporated into a balanced, semisynthetic diet so as to supply 20% protein. A diet of the same composition but containing casein as the protein source was used as the control and a practical laboratory chow was used as a standard reference diet.

Diets were fed to weanling Sprague-Dawley rats (Charles River Breeding Laboratories, Wilmington, Massachusetts), thirty males and thirty females assigned to each diet. Food intakes were recorded daily, and each animal was visually examined daily and weighed weekly.

Rats from each group were bred during the test period, and the second litter from each female used for further generation studies or for measurements of growth, reproduction, lactation, feed efficiency, haematological urinary or histopathological evaluation. Offspring from all groups were examined for abnormalities at birth.

The casein control diet contained less than 0.01 p.p.m. total mercury by analysis, and a sample of rat chow diet contained 0.06 p.p.m. of total mercury. All lots of FPC were analysed for total, inorganic, and methyl mercury according to methods of Westoo and Rydall⁹ in the laboratory of Dr T. W. Clarkson, University of Rochester. Results are shown in Table 1. The average total mercury content of the FPC samples was 0.22 p.p.m., with more than 90% as methyl mercury.

Typical body weights over the 2-yr period in the F₁ generation and 28-day food efficiency in F₁ and F₃ generations indicated that rats fed FPC did as well as or better than those fed casein or chow, and there was no evidence of a toxic response. Furthermore, in a 28-day study, weight gain and food efficiency in the FPC group were found to be equal to or better than either of the control groups. Table 2 illustrates characteristic reproduction performance. Judging from numbers weaned in

Table 1 Total Inorganic and Methyl Mercury Contents of Fish Protein Concentrate Samples

Sample No.	Total mercury (p.p.m.)	Inorganic mercury (p.p.m.)	Methyl mercury (p.p.m.)
146	0.21	<0.01	0.26
147	0.33	<0.01	0.20
149	0.23	<0.01	0.22
151	0.21	<0.01	0.17
153	0.17	<0.01	0.16
154	0.18	0.02	0.18
155	0.17	<0.01	0.15
156	0.30	0.03	0.28
Average	0.22	<0.01	0.20

Analyses were performed by Dr T. W. Clarkson, School of Medicine and Dentistry, University of Rochester, Rochester, New York.

Table 2 Reproduction Studies, F₁ and F₃ Generations

	No. litters		Average No. pups per litter		Average weight pups	
	F ₁	F ₃	F ₁	F ₃	F ₁	F ₃
FPC	27/30	26/29	9.0	9.5	6.1	6.0
Casein	21/30	24/29	9.4	9.7	6.0	6.2
Chow	26/30	29/29	11.2	12.2	6.1	6.0

second litters of each group, performance in the FPC group was as good as or better than either casein or chow groups. The average number of pups was equal in FPC and casein groups, both of which had fewer on the average than the chow group. Birth weights were the same for all three groups. These observations further support data which indicate that FPC containing low levels of mercury had no adverse effects on conception and pregnancy. Congenital anomalies or abnormalities were not observed in any of the litters of any of the generations, and at no time in any group were significant clinical signs or symptoms noted. Results from F₂ and F₄ generations are not shown since nothing unusual was found in either.

Furthermore, all values for haematocrit, white and red blood cells, differential, white cells, haemoglobin, and all urinalyses (creatinine, creatinine, protein, specific gravity, osmolality) were within normal ranges for all groups. Detailed histopathologic studies revealed no lesions in the brain, liver or kidney attributable to diet; the only lesions observed were associated with age and were essentially the same for all groups.

Samples of liver, brain, and kidney from the F₁ generation, fed FPC for 2 yr, and F₃ generation, fed FPC for 1 yr were analysed for total mercury⁹. Results of analyses from F₁ are shown in Table 3. There were no significant differences between F₁ and F₃ generations. Mercury was not detected in the livers and brains of animals fed casein and very little was detected in the kidneys. Mercury was detected in all tissues of animals fed rat chow, with most found in the kidney. Rats fed FPC diets accumulated more mercury in all tissues than the other two groups, with considerably more in the kidneys than in the liver and brains. Females tended to accumulate more mercury than males. Most reports^{4,9} indicate that methyl mercury tends to accumulate in the central nervous system. Although the mercury content of the FPC used in this study was largely in the methyl mercury form, most of that in the rat tissues was in the kidney with very little in the brain. We did not assay for the various forms in the tissues, however. This points out the need for more data from many different laboratories before generalizations can be made on the form and location of the different mercury compounds in biological systems.

In spite of the greater amounts of mercury in the tissues of animals fed FPC diets, no gross, histologic or other untoward effects were noted in the rats, the growth and reproductive performance of which were comparable with the performance of those fed casein or chow. Thus, the amounts of mercury

Table 3 Tissue Total Mercury Content of Rats Fed Three Diets, F₁ Generation (expressed as p.p.m., Wet Tissue)

	Liver F ₁		Brain F ₁		Kidney F ₁	
	M	F	M	F	M	F
FPC						
Mean	0.22	0.24	0.08	0.12	1.38	2.12
Range	0.19-0.27	0.16-0.37	0.05-0.10	0.08-0.17	1.11-1.73	1.48-2.58
Casein						
Mean	0	0	0	0	0.05	0.05
Range	0	0	0	0	0.02-0.07	0.03-0.07
Chow						
Mean	0.03	0.06	0	0.03	0.18	0.44
Range	0-0.08	0.03-0.09	0-0.03	0-0.04	0.02-0.38	0.25-0.56

consumed by the rats fed FPC had no detectable toxic effect or influence on longevity or reproduction.

These findings are particularly significant in relation to the question of the amount of mercury that can be consumed without hazard to human health. In these studies, FPC was included in the diet at a level of about 25%. Since one unit of FPC represents about six units of whole fish, considerably more fish in the form of FPC was used in the diet than would have been possible through the use of fresh fish. 25% FPC (or even fresh fish) in the diet is abnormally high compared with the amount of fish in normal human diets.

The FPC used in these studies contained an average of 0.22 p.p.m. of mercury. Diets containing large amounts of FPC, however, raised the average total consumption of mercury by the rats to be about 12 µg/kg of body weight per week. Extrapolated to a 70-kg man, this intake is equivalent to 840 µg of mercury per week. If this amount of mercury were supplied by fresh fish containing 1.0 p.p.m. of mercury (two times the FDA action level), a total consumption of 1.85 lb of fish would be required per week or about 96 lb per year. This amount is more than eight times the average amount of fish per capita that has been consumed each year in the United States for the past 50 years.

The results from these studies do not indicate the maximum amount of mercury that can be consumed without hazard. The rat, however, may not be the best model for providing data for extrapolation to man¹⁰⁻¹².

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Polychlorinated Biphenyls in Carbonless Copying Paper

POLYCHLOROBIPHENYLS (PCB) cause chloracne and liver damage in man and induce oestradiol degradation¹. They are widely distributed in the ecosystem in birds, fish and shellfish¹⁻⁴. The miserable episode of "Yusho", a mass food poisoning involving more than 1,000 people which was caused by chronic ingestion of rice oil contaminated with 'Kanechlor-400', a commercial brand of PCB, occurred in western Japan in 1968^{5,6}.

Because of their unique physical and chemical characteristics, PCBs are widely used in industry as heat transferring media, electric insulators, lubricants, plasticizers and so forth. PCBs are also used for manufacturing carbonless copying paper, the use of which has rapidly increased in Japan in recent years.

Chemical analysis was therefore carried out for three sheets of each of three Japanese brands of such paper. These sheets were extracted separately with acetone in a soxhlet apparatus for 3 days until there were virtually no gas chromatographic peaks. Each extract was subjected to gas chromatography using 'Shimadzu GC-5A' with a detector of nickel 63 electron capture and three different liquid phases—5% SE-30, 5% QF-1, and 2.5% DEGS—coated on 'Chromosorb W AW DMCS'. The gas chromatographic patterns were identical to those of 'Kanechlor-300' (chlorine content 43%) (Fig. 1). The infrared absorption spectra of the extracts demonstrated the presence of 'Kanechlor-300' together with paraffin. The fragmentation pattern of mass spectrum of the evaporation residues was very similar. The presence of biphenyls possessing five chlorine atoms was indicated by the existence of the group of highest mass starting from *m/e* 324. The approximate concentration of PCB in the paper, as determined by gas chromatographic peak heights which linearly increased up to 40 ng of 'Kanechlor-300', is shown in Table 1, 2-6% in the upper and middle sheets and 0.02% in the lower sheet.

To estimate how much PCB sticks to fingers in the handling of the papers, we counted three sets of the upper, middle, and lower sheets, each consisting of thirty-two pre-cut sheets of the same size, by turning the sheets over one by one. The fingers were then washed with 20 ml. of n-hexane once, the washings concentrated to a small volume (1-4 ml.) and their PCB contents determined by gas chromatography. The amounts of PCB in

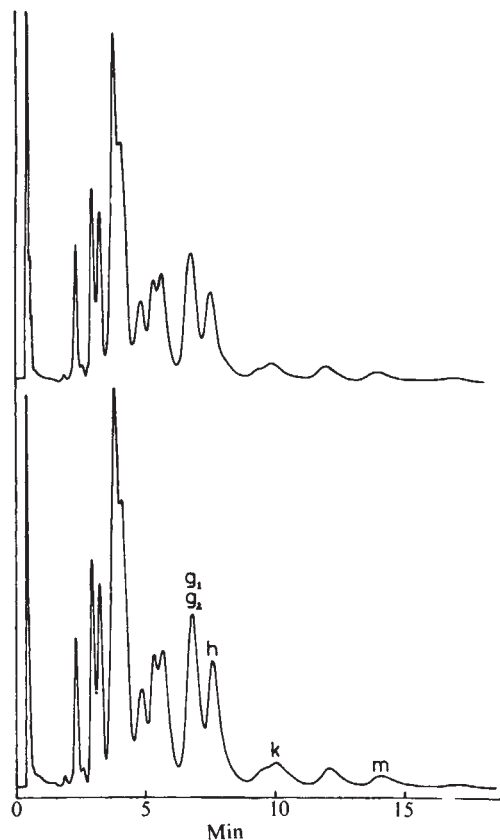


Fig. 1 Gas chromatograms of 'Kanechlor-300' (upper) and the acetone extract of copying paper (lower). Glass column (3 mm $\times$ 2 m) containing 5% SE-30 on 'Chromosorb W AW DMCS' at 195° C.

the washings ranged from 11.40 to 52.68 μ g and 30.22 μ g on average, in five subjects (Table 2). A repetition of this experiment in which the hands were washed with soap after the counting and before hexane washing showed that only one-third of the PCB could be removed by ordinary hand washing. It is therefore possible for the substance to be absorbed through the skin unless special precautions are taken.

Table 1 Amounts of PCB in Carbonless Copying Paper

Brand	Kind of paper	Weight of paper (g 100 cm ⁻²)	Amount of acetone extract* (mg g ⁻¹)	Amount of PCB found† (mg g ⁻¹)
A	Upper	0.47	117.0	64.7
	Middle	0.56	125.8	63.8
	Lower	0.51	23.4	0.24
B	Upper	0.48	40.8	30.7
	Middle	0.55	37.8	26.0
	Lower	0.51	79.8	0.28
C	Upper	0.40	59.6	31.6
	Middle	0.53	61.0	22.2
	Lower	0.50	28.2	0.20

* Weight of extract per weight of paper.

† Weight of PCB calculated as 'Kanechlor-300' per weight of paper.

According to the manufacturers and also our scanning electron microscopy of the paper surface (courtesy of Dr Y. Ishii), the under-surfaces of the upper and middle sheets are coated with gelatine capsules containing dye, for which PCB seems to be used as the solvent.

The PCB found in the papers was 'Kanechlor-300', which is

Table 2 Amounts of PCB Found from Fingers after Counting Ninety-six Sheets of Paper

Subject	Amount of PCB*	
	Washed with n-hexane (μ g)	Washed with n-hexane after hand washing with soap (μ g)
1	29.76	16.39
2	12.60	10.47
3	11.40	3.50
4	52.68	47.15
5	44.68	25.45
Average	30.22	20.60
Standard deviation	16.60	15.11

* Calculated as 'Kanechlor-300'.

43% chlorine, and not 'Kanechlor-400' which is 48% chlorine and caused the incident of "Yusho". There is no doubt that they have many common components. For highly chlorinated components, however, the peaks designated g₁, g₂, h, k, and m in the gas chromatogram (Fig. 1) coincide with 2,4,3',4', 2,5,3',4', 2,3,4,4', 2,4,3',4'-tetrachlorobiphenyl and 2,3,4,3',4'-pentachlorobiphenyl respectively which were identified in 'Kanechlor-400' by Saeki *et al.*⁷

Because highly chlorinated PCB, once absorbed, easily accumulates in the adipose tissue and is hard to detoxicate or eliminate, we consider that its use for carbonless copying paper should be discontinued.

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Mortality of Thick-billed Murres in the West Greenland Salmon Fishery

SINCE its inception in 1965, the drift net fishery for Atlantic salmon (*Salmo salar*) off West Greenland has taken a heavy toll of thick-billed murres (*Uria lomvia*)¹, which are entangled and drowned as they dive for food.

The commercial drift net fishery is situated in the offshore waters along the west coast of Greenland, between 60° and 70° north latitude, from August to December, with the peak catch in September and October. The nets float at the surface with a depth of 6 to 10 feet, set in a line which may be up to 10 miles long. In this communication we list the seabirds drowned in nets set by the Fisheries Research Board of Canada trawler A. T. Cameron in the autumns of 1969, 1970 and 1971, during

salmon-tagging operations. We estimate the total yearly kill in the commercial drift net fishery, and couple this with population estimates and other mortality data to show that the annual kill of thick-billed murres in the North-West Atlantic has become excessive and cannot be tolerated by the populations involved.

To estimate the total annual murre kill we have grouped the data from each square of one degree of latitude by one degree of longitude within the region of the commercial fishery. We calculated the numbers of salmon and murres caught per unit fishing effort for each one degree square. ("Fishing effort" is the number of nautical miles of drift net which were set, multiplied by the number of hours during which they were set.) We did this for each year separately, and for all 3 years combined. Figures for the average number of salmon and murres caught per unit fishing effort during the whole region were obtained by weighting each one degree square equally and then averaging all the figures for a given year, or for all 3 years combined. (It would have been preferable to have weighted the data according to the relative importance of various areas to the commercial fishery, but detailed information on this is not available.) The ratio of murres caught to salmon caught was then calculated. On the assumption that the ratio obtained in experimental fishing would also apply to the commercial fishery, this was applied to the total salmon catch to obtain an estimate of the total murre catch.

and 540,000 using the ratio obtained from the combined data of all 3 years.

The variation in estimates shows that there is a large margin for possible error in the above extrapolation to a total murre kill. One reason may be the small sampling by the research vessel which has provided the only recent quantitative data. Moreover, the 1969 sets were chiefly in Disko Bay where there is very little commercial drift net fishing. Also much of the 1969 fishing was during daylight hours as opposed to the commercial practice of fishing overnight, whereas many of the sets of 1970 and 1971 were put out at night. The evidence suggests that most of the murres are caught at night. The 1970 and 1971 estimates are probably more representative of the actual situation than is that for 1969. But it is clear that the yearly kill is in the hundreds of thousands.

We therefore feel that a kill of the order of 500,000 murres yearly is a reasonable estimate, which may however, be in error by as much as $\pm 50\%$. We should stress that this applies only to 1969, 1970, and 1971 when the commercial drift net catches were of the order of 1,200 metric tons. The 1968 commercial drift net catch was only 548 metric tons², so the estimated murre kill should be reduced accordingly.

The only previous extrapolation of a total murre kill¹ is erroneous. It calculated the ratio of murres to salmon caught (0.88) by the single drift net vessel which fished the West Greenland area in 1965. This ratio was then applied to the

Table 1 No. of Sets, Salmon Caught per Unit Fishing Effort (s/e), and Murres Caught per Unit Fishing Effort (m/e) for each Square of 1° of Latitude by 1° of Longitude

Square °N × °W	No.	1969 s/e	m/e	No.	1970 s/e	m/e	No.	1971 s/e	m/e	No.	3 yr s/e	m/e
69 55				2	0.61	0.27				2	0.61	0.27
69 52	10	1.72	0.02				2	0.43	0.02	12	1.53	0.02
67 55				1	0.76	0.08				1	0.76	0.08
67 54				2	0.43	0.04	5	2.09	0.94	7	1.52	0.63
66 55				2	1.18	3.27				2	1.18	3.27
66 54	4	0.41	1.63	2	1.06	4.24				6	0.67	2.66
65 53	1	0.41	0.00	1	0.75	4.52	1	2.34	0.05	3	1.29	1.54
64 53				2	0.77	0.42				2	0.77	0.42
63 52							1	0.33	4.45	1	0.33	4.45
Average		0.85	0.55		0.79	1.83		1.30	1.36		0.96	1.48
Murres/salmon		0.65			2.32			1.05			1.54	

Within the region of the West Greenland salmon fishery, the A. T. Cameron in 1969 caught 598 salmon, 177 murres (undifferentiated as to species but undoubtedly almost entirely thick-billed murres), two dovekies (*Plautus alle*), and two black guillemots (*Cepphus grylle*) in fifteen drift net sets (September 15 to October 2). In 1970 the catch was 257 salmon, 560 thick-billed murres, one common murre (*Uria aalge*), one dovekie, and one common puffin (*Fratercula arctica*) in twelve sets (September 3 to September 25). In 1971 the catch was 363 salmon, 246 thick-billed murres, two common murres and two black guillemots in nine sets (September 6 to September 16). The thick-billed murre is thus the only species of seabird being taken in large numbers in the drift net fishery.

The ratios of murres to salmon caught in the experimental fishing were 0.65 for 1969, 2.32 for 1970, 1.05 for 1971, and 1.54 for the combined data of all 3 years (see Table 1). The 1969 commercial drift net catch was approximately 353,000 salmon (assuming an average fresh weight of 3.5 kg)². Commercial drift net catches for 1970 and 1971 are not known precisely but are of the same order as is that of 1969. Using the 1969 commercial drift net catch as a basis, the annual murre catch is estimated to be 230,000 based on the 1969 murre/salmon ratio, 820,000 on the 1970 ratio, 370,000 on the 1971 ratio,

1964 salmon catch, although this was obtained by means of shore gill nets only. The resulting estimated murre kill for 1964 is meaningless. The ratio obtained is, however, consistent with those obtained in this work.

Banding returns indicate that the kill affects chiefly murres from colonies in West Greenland and the eastern Canadian Arctic³⁻⁵; there may also be some birds from East Greenland and Spitzbergen⁵. Population estimates of thick-billed murres in this region are very uncertain. Salomonsen (personal communication) in 1970 estimated the West Greenland population to be 1,500,000 breeding pairs, while Tuck³ in 1960 estimated the eastern Canadian Arctic population to be 2,000,000 breeding pairs. About forty-one thick-billed murre chicks go to sea per 100 breeding pairs³, or approximately 1,500,000 chicks from all the West Greenland and Canadian Arctic colonies.

The various mortalities to which the murres are subjected can now be shown to be excessive. Salomonsen⁶ gives a yearly hunting toll of 750,000 thick-billed murres in West Greenland, where this is the most important bird species in the economy. Thus the two mortalities caused by man for which figures are available, from the drift net fishery and from hunting in West Greenland, approach the yearly production of chicks in the

region. No account has been taken of the natural pre-breeding mortality of the young once they reach the sea, and of the natural mortality of the adults. There are no documented estimates of the mortality due to oil pollution (known to be heavy in the wintering areas off Newfoundland, L. M. Tuck, and R. G. B. Brown, personal communication), and of the winter hunting toll of murres in Newfoundland waters (of less importance than the oil pollution mortality, L. M. Tuck, personal communication).

When deaths from all causes are considered, the annual mortality of thick-billed murres in the western North Atlantic exceeds the annual production of young. Steps should be taken to reduce these mortalities caused by man, especially the high tolls in the drift net fishery and by hunting in West Greenland. Action should also be taken toward a better utilization of the birds caught in the drift nets in order that the hunting toll could be correspondingly reduced. Unless such measures are taken, the future of the thick-billed murre in this region is subject to grave doubt.

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Origin of Plastids and the Phylogeny of Algae

THERE have been many suggestions that chloroplasts are derived from endosymbiotic prokaryotes¹⁻⁴. All the proposed evolutionary schemes, with the exception of Raven's⁵, have avoided the question of exactly where the transition from the prokaryotic to the eukaryotic condition in plants occurred⁶⁻¹³. In this article I will discuss Raven's hypothesis (Fig. 1) and present an alternative hypothesis (Fig. 2).

Any evolutionary scheme should adhere to the following three principles. (1) A monophyletic origin of any organism, chemical compound or cytoplasmic structure has the greatest statistical probability of being correct. (2) The loss of a non-essential structure can require just the mutation of a single gene but the acquisition of a structure generally requires many mutations and a considerable amount of time. (3) Most

organisms in evolutionary sequences would have been lost, yet in postulating phylogenetic events the plausibility of the theory can be enhanced by the existence of organisms similar to those in the proposed scheme.

Raven⁵ concludes that there were originally three prokaryotic groups (Fig. 1); the blue-green algae, the "green prokaryotes" and the "yellow prokaryotes".

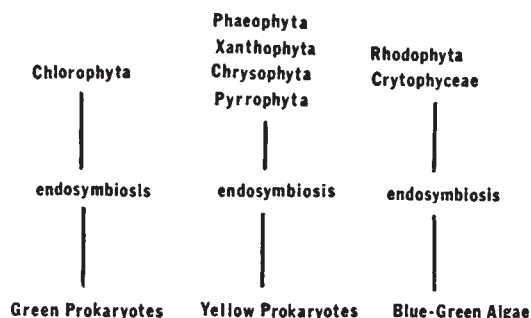


Fig. 1 Multiple origin of plastids as proposed by Raven. Three prokaryotic groups became involved in separate endosymbioses to yield three different eukaryotic groups of algae.

Each prokaryotic group became involved in an endosymbiosis which subsequently evolved into the chloroplasts of a group of eukaryotic algae. The primary advantage of this theory is that the pigments of the supposed prokaryotic groups are the same as their eukaryotic counterparts. The blue-greens, Rhodophyta and Cryptophyceae, have chlorophyll a and phycobilins in common. The "yellow prokaryotes" and the Chromophyta⁴ minus the Cryptophyceae have chlorophyll a and c. The "green prokaryotes" and Chlorophyta have chlorophyll a and b.

There are some serious disadvantages to Raven's theory, however. (1) There are no existing organisms or endosymbioses similar to the hypothetical "yellow" or "green prokaryotes" nor is there any evidence that there ever were such organisms. (2) Many of the morphologically simple and probably primitive algae in Raven's three derived eukaryotic groups contain pyrenoids. Raven does not explain the origin of the pyrenoid and, since the pyrenoid structure in all algae is similar, it is necessary in his scheme to derive an identical structure along three different phylogenetic lines. (3) Two of Raven's eukaryotic groups have chlorophyll c, the Cryptophyceae and the remaining groups in the Chromophyta. The presence of chlorophyll c in both of these groups is stated as a result of separate evolution. (4) Some members of all three eukaryotic groups have chloroplast endoplasmic reticulum¹⁴, which in Raven's theory requires a triphyletic derivation of the same condition. (5) The Prasinophyceae of the Chlorophyta and the Cryptophyceae have ejectosomes of similar structure^{16,17} necessitating a diphyletic derivation in Raven's theory. (6) *Cyanophora paradoxa* is a Cryptophycean alga with endosymbiotic blue-green algae which has characteristics of the Cryptophyceae and Dinophyceae¹⁸. Such a relationship would be difficult in Raven's scheme because the Cryptophyceae and Dinophyceae are the result of two separate evolutionary lines.

If the extant eukaryotic organisms are surveyed for endosymbioses, the only prokaryotic organisms in such associations are Cyanophyceae¹⁹. The relationship between an endosymbiotic Cyanophycean alga and its host is a syncyanosis, the endosymbiotic Cyanophycean alga is a cyanelle and the cyanelle plus the host organism is a cyanome²⁰.

Plant syncyanoses can be divided into four groups¹⁹: Cryptophyceae (three genera), Glaucophyta²¹ (five genera), Bacillariophyceae (one genus), Phycomycetes (one genus). If most of these organisms had a single ancestral type there is a

good possibility that it was in this that the evolution of chloroplasts occurred.

The Glaucophyta are the key as to the group of organisms in which chloroplasts evolved. The Glaucophyta have reserve product produced in the cytoplasm of the host, phycobilin pigments in their cyanelles and at least one of the Glaucophyta, *Glaucocystis*, has two rudimentary flagella inside the cell wall²²⁻²⁵. The only class of flagellated organisms with phycobilins and reserve product in the cytoplasm is the Cryptophyceae. It has been indicated that the Glaucophyta are advanced types of syncyanoses and that the establishment of the syncyanosis in present conditions is impossible because of the rigid cell walls of the organisms^{21,25}. At the time that the original symbiosis was established, therefore, the ancestor of the Glaucophyta probably lacked a cell wall, had two functional flagella and was actually a Cryptophycean alga. If the Glaucophyta are considered to be of Cryptophycean ancestry then eight of the ten genera that have cyanelles are Cryptophyceae or of Cryptophycean ancestry. Such evidence points to the evolution of the chloroplast in a Cryptophycean alga. The Cryptophyceae also have the necessary sequence for a group involved in the evolution of a chloroplast from a Cyanophycean alga. The Cryptophyceae have colourless, phagocytic organisms with gullets, organisms with cyanelles and organisms with true chloroplasts.

If the cyanelles of the Cryptophyceae evolved into chloroplasts then a cytological structure that might serve as a precursor to the pyrenoid should be present in these Cryptophycean cyanelles, and also possibly in those of the Glaucophyta. The cyanelles of the Cryptophyte *Cyanophora paradoxa* have a crystalline central body²⁶ surrounded by DNA microfibrils. The pyrenoid of the Dinophyceae and the Bacillariophyceae has a crystalline matrix^{27,28} similar to that of the central body of *Cyanophora*. The evolution of the cyanelle of *Cyanophora* into a chloroplast would result in the central body developing into a central pyrenoid. *Glaucocystis nostochinearum* has a lamella-free area in its cyanelles that has a crystalline matrix²⁴ similar to that in the central body of *Cyanophora*.

characteristics in common; gullets, muciferous bodies, solid reserve product outside the chloroplast, flagella with mastigonemes, chloroplast endoplasmic reticulum, para-basal dictyosomes, no cell walls^{15,18,30-32}. The Prasinophyceae and the Cryptophyceae have ejectosomes in common^{16,17}. The Prasinophyceae and the Chlorophyceae have starch produced within their chloroplasts^{14,33} which means that at some time during their evolution the cyanelles or the chloroplasts must have taken over storage of the reserve product from the host cytoplasm (probably in the more primitive Prasinophyceae³³).

The second phylogenetic line from the Cryptophycean cyanome required the development of chlorophyll c from chlorophyll a by the loss of the phytol tail of chlorophyll a²⁹. Once chlorophyll c evolved the cyanelles evolved along two lines, the first to the chloroplasts of the Cryptophyceae and the second to the chloroplasts of the remainder of the Chromophyta (requiring the loss of phycobilin pigments). The Cryptophyceae and the remainder of the Chromophyta have many characteristics in common; chlorophyll a and c¹, at least one flagellum with mastigonemes³⁴, chloroplast endoplasmic reticulum, para-basal dictyosomes, reserve product produced outside the chloroplast¹⁵. In addition the Cryptophycean cyanome, *Cyanophora paradoxa*, has plates in the peripheral cytoplasm that are encased in vesicles with subtending microtubules¹⁸. This condition is characteristic of the Dinophycean³⁵ and *Cyanophora* probably represents a transitional stage between the two classes.

The third phylogenetic line, from the Cryptophyceae containing chloroplasts to the Rhodophyta, required the loss of flagellation and chlorophyll c and the acquisition of a cell wall. The derivation of the Rhodophyta from a flagellated ancestor has been generally discounted in modern papers on algal phylogeny⁹⁻¹³. Some authors^{12,13} have stated that because the Rhodophyceae lack centrioles or basal bodies they did not have a flagellated ancestry. Pickett-Heaps³⁶ has pointed out that centrioles have nothing to do with the actual division of the cell and are merely "along for the ride". If a flagellated alga is in an environmental position where it would survive as well without flagella, then an abortive division of the centriole with the resulting loss of flagellation, could be the beginning of a new non-motile evolutionary line.

The advantages of the above phylogenetic system are that no multiple derivations or sudden syntheses of cytoplasmic structures or chemical compounds are required. Also the proposed pathways can be partially substantiated by extant remnants of the populations that participated in the evolutionary process.

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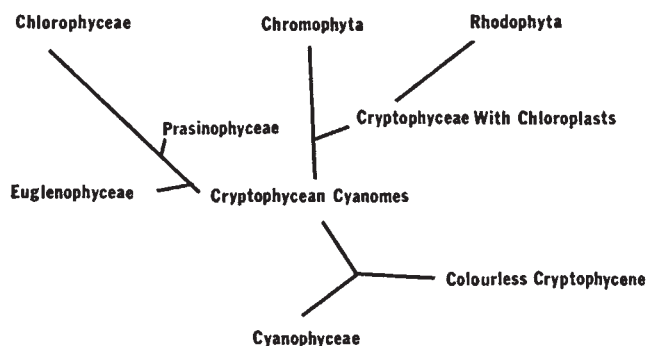


Fig. 2 The origin of plastids from a Cyanophycean alga involved in an endosymbiosis with a colourless Cryptophyte. The remainder of the eukaryotic algae then evolved from this Cryptophycean cyanome (endosymbiotic Cyanophycean alga plus the host cell).

The development of the eukaryotic algae from the cyanomes of the Cryptophyceae probably occurred along three separate lines (Fig. 2), the first to the Chlorophyta, the second to the Chromophyta and the third to the Rhodophyta.

The first line of evolution from the Cryptophycean cyanome was to the Chlorophyta. This evolutionary sequence involved the loss of phycobilin pigments and the acquisition of chlorophyll b (basically requiring the addition of one aldehyde group to chlorophyll a²⁹).

The Cryptophyceae and the Euglenophyceae have many

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Nickel Pollution

IN view of the present concern about pollution of the environment by airborne metals¹, the following evidence of contamination of herbage and soils on a farm in South Wales in 1934-36 may be of interest². The farm was situated near Clydach about 1 mile north-east of a large nickel works which also produced some copper, and was therefore subject to contamination from prevailing winds. Samples of herbage and soil were sent to the Department of Agricultural Chemistry at Aberystwyth by a veterinary surgeon who was concerned about the poor condition of livestock.

Qualitative examination showed the presence of considerable amounts of nickel with smaller amounts of copper in both herbage and soil. Pasture herbage here had a "burnt" appearance, and consisted chiefly of fescues and bent which formed a loose mat easily dislodged and suggesting poor root development. Trees were withered and appeared to be dying.

Table 1 Percentage of Nickel in Herbage Dry Matter and Air-dried Soil

Grass samples										Soil	
1	2	3	4	5	6	7	8	9	Con- trol*	1	2
0.17	0.18	0.17	0.22	0.18	0.22	0.17	0.22	0.32	Nil	0.05	0.06

* Herbage sample from college farm.

Sufficient nickel was present in HCl extracts of ashed herbage and air-dried soil to allow gravimetric determination by the well known dimethylglyoxime procedure³ (Table 1). All samples, which were taken during the summer of 1934, contained small amounts of copper, and the soils gave definite positive reactions for arsenic with traces only in the herbage.

The levels of nickel represent gross contamination (1,700 to 3,200 p.p.m. compared with levels of 6-82 in *Festuca rubra*

found by Goodman and Roberts¹), and we determined whether it appeared mainly on the surface or within the plant. Samples of wheat and oats were separated as shown in Table 2 and some leaves washed in running water before analysis. Thus, leaves which expose the largest surface and emerge before the heads contained by far the largest amounts of nickel, whereas straw encased by the leaf sheath contained traces only. Moreover, washing removed most of the nickel from the leaf surface. Further evidence of surface contamination was obtained by analysing "winter burnt" herbage exposed to contamination throughout the winter and young growth from a 1 yr old ley sampled in March 1936. The dry matter of these contained 0.36-0.41 and 0.09% of nickel respectively. Only minute traces of nickel and copper were found in washed peeled potatoes.

Table 2 Localization of Nickel in Oats and Wheat

Sample		Nickel (p.p.m. of dry matter)
Oats 1	Leaves	230
	Heads	60
	Straw	Trace
Oats 2	Leaves	230
	Heads	70
	Straw	Trace
Oats 3	Leaves	250
	Heads	80
	Straw	Trace
Oat leaves from a rural area		Nil
Wheat leaves	Unwashed	250
	Washed	70
Unripe wheat	Heads	80
	Chaff	80
	Grain	Trace

It was found that approximately 20% of the nickel deposit was dissolved when grass containing 0.36% nickel was incubated at 37° C for 24 h with 500 ml. 1.25% HCl containing 1 g of pepsin (a procedure similar to that used to determine *in vitro* digestibility of herbage protein at the time).

Six samples of vegetation and soil taken on September 20, 1971, were analysed by atomic absorption spectrophotometry and found to contain nickel as follows (p.p.m. Ni on a dry weight basis) (Table 3). Samples 1 and 2 consisted of nardus and heather respectively growing on moorland, where *a* and *b* represent the first and second inch of topsoil, the former rather peaty. Sample 3 was lush grass aftermath from a ley whose soil would have been disturbed by ploughing, while 4 and 5 were nardus and birch leaves from a slope facing the works where herbage was sparse and brown, and soil very poor. Sample 6 was bracken growing along the roadside near samples 1 and 2.

Table 3 Atomic Absorption Spectrophotometry of Soils and Vegetation

Sample	1	2	3	4	5	6
Vegetation	1.3	1.4	1.0	3.4	4.5	2.0
Soil <i>a</i>	625	350	1,150	—	—	—
<i>b</i>	200	350	350	—	—	—

The marked decrease in contamination of the vegetation since the 1930s is probably due to the adoption of a much cleaner process of nickel extraction after 1940. The high levels in topsoil merit further attention (precipitates were obtained

with dimethylglyoxime). In taking soil samples in 1934, no attempt was made to obtain surface soil.

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Analysis of Distress Calls of Chicken × Pheasant Hybrids

THE inheritance of some simple behavioural traits, according to Scott and Fuller, can be explained on the basis of one or two genes¹. Hirsch has suggested that it is impossible to study the genetics of a behaviour and that only the genetics of a population can be analysed, on the grounds that the relation between behavioural variation and relevant genetic variation is never constant². We believe that "fixed action patterns" (*Erbkoordination*³) of behaviour are accessible to genetic analysis if genetic differences are demonstrable for the parameters of those behaviours in the parents.

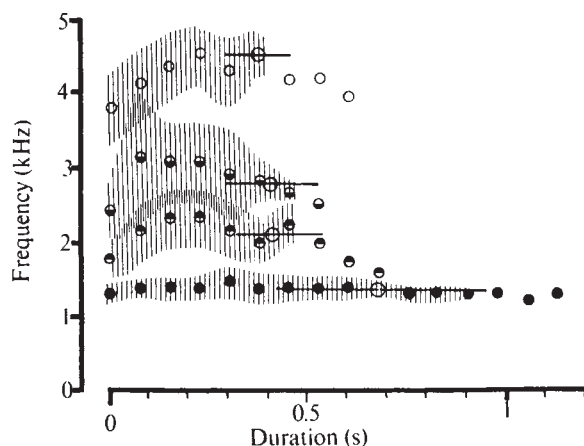


Fig. 1 Means and standard deviations for the frequency (○) and duration (◐) of the principal component of hand-held distress calls. The durations for the chicken (male and female) include only the 70% of the call showing a measurable frequency component. Standard deviations for the last points were not calculated because of the low number of calls of those durations. ○, Pheasant; ◐, small hybrid; ◑, large hybrid; ●, chicken.

Previous studies of hybrid birds have revealed behaviour patterns either intermediate to those of the parental species, similar to one or the other parental species, or not present in either parental species and possibly suggestive of behavioural atavism⁴⁻⁷. We have investigated the effects of the genotype on a relatively simple behaviour pattern, the distress call emitted by hand-held birds. Such distress calls⁸ of white Leghorn chickens (*Gallus domesticus*)⁹, ring-necked pheasants (*Phasianus colchicus*) and their hybrids were recorded with a Uher model 4000 Report-L tape recorder and a Uher model M514 microphone with a frequency response range of 40–16,000 Hz at 3.75 i.p.s. in an anechoic studio. The calls were

analysed using the Kay Electric Company Sona-Graph 6061A at the wide band (300 Hz) setting.

White Leghorn semen was used for artificial insemination of pheasant hens to produce the hybrids at the US Department of Agriculture Research Center, Beltsville. The rearing environment of the birds was such that the possible influence of learning cannot be firmly established. The work of Schjelderup-Ebbe¹⁰ and Konishi¹¹, however, indicates that for the domestic fowl learning plays little if any role in the development of the vocal repertoire. The sex of the hybrids was determined by dissection or chromosome karyotypes (P. S., unpublished). The 1 yr old hybrids used in this study showed two size classes, the small birds, females, having a mean weight of 875 g and the large birds, males, having a mean weight of 1,755 g. The mean weights of the male chickens and female pheasants were 2,380 g and 1,025 g respectively.

Five males and five females of each parental species, five small hybrids, and six large hybrids were subjected to the test situation. Each group of birds was acclimated to the anechoic studio for 22 h before the recordings of the distress calls which were taken between 1000 h and 1200 h. Birds were individually netted, removed from the net and hand-held with wings dorso-medially. Five-minute recordings were obtained from each bird within the studio for each of five trials, thereby providing 20-min intervals between successive trials for each bird. This procedure ensured relative uniformity of motivational states of the animals. Fig. 1 represents the means and standard deviations of frequency and duration of the principal component of the distress calls for each bird group. The principal component was operationally defined as that harmonic which appeared consistently darkest and most distinct in the sonagram.

Table 1 Duration of Calls of the Chicken × Pheasant Hybrids and their Parents

	<i>n</i> calls	Mean (s)	s.d. (s)	V
Pheasant	34	0.38	0.07	0.18
Small hybrid	41	0.41	0.12	0.29
Large hybrid	69	0.42	0.13	0.31
Chicken	44	0.68	0.27	0.40

There was no obvious difference between the distress calls of the male and female chickens. The male pheasants, however, did not emit a distress call in the test situation, whereas four of the five female pheasants did (Fig. 1). The intraindividual and interindividual variations of the distress calls within a species or hybrid size class were not significantly different, so that data from all individuals of one class were pooled. The principal component of the chicken calls showed little change in frequency from beginning to end, with about 30% of the call lengths comprising initial "noise", as defined by Busnel to be "sound without regularities, confused sum of many unrelated sounds heard together"¹². The principal component of the female pheasant calls showed initial rise in frequency, as well as a sharp terminal drop in frequency encompassing about 1,500 Hz.

The means of frequency of the principal components of both hybrids' distress calls were intermediate between those of the parent species. Frequency modulation ranging from 50–80 Hz was present in the calls of the chicken and both hybrid types, but was absent from those of the pheasant. Noise was unevenly spaced but pervasive in the pheasant and hybrid calls. The mean duration of the calls of both hybrid groups was similar to that of the pheasant as shown in Table 1. Further, an initial rise in frequency of the principal components of the calls was noted in hybrids and pheasants, but was absent from the chicken calls. Certain hybrids of both size classes, however,

resembled more closely either the chicken or the pheasant with respect to general harmonic structure.

Analysis of the calls suggests that the inheritance of the frequency of the principal call component may be polygenic, whereas the duration of the call may be determined by one or a few genes. These statements should not be considered as more than speculative, however, for evidence from backcrosses is not available.

The data show that scrutiny of the parameters of a simple behaviour pattern, rather than the whole behaviour pattern itself, may lead to a more productive approach to the investigation of behaviour genetics. This approach is supported by the fact that the parameters of a simple behaviour pattern such as the frequency and duration of the distress call seem to be genetically controlled in different ways.

Continuing studies of the social and vocal behaviour of the chicken $\times$ pheasant, as well as chicken $\times$ Japanese quail (*Coturnix coturnix japonica*) and pheasant $\times$ Japanese quail hybrids¹³ should reveal the patterns of inheritance of individual elements of fixed action patterns.

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Territorial and Courtship Songs of Birds

MORSE¹ has postulated that the two song types of the black-throated green warbler (*Dendroica virens*) have different functions: song A for territorial defence and song B for courtship and pair-bond maintenance. On the basis of several years of study of the songs of this and other species of Parulidae, I feel it is possible to criticize Morse's hypothesis on several points and to suggest an alternative explanation.

"Causation" of bird song can be discussed in three senses: that of the evolutionary function for which the communication pattern has been selected; the hormonal-neurological basis; and the external stimuli which determine the type of song and the rate of delivery. Morse has attempted to draw conclusions about the evolutionary "causation" of bird song from observations of the stimuli which induce it.

The assumption of separate functions for the two song types can be questioned on a theoretical basis. Smith² has shown how the same "message" from a signalling bird may have different "meanings" depending on the context and the status (sex, age,

territorial status and so on) of the recipient. The message represented by either song type of *D. virens* could, and very probably does, therefore, have different meanings for the males and for the females of that species. If the repertoire of a species is limited^{3,4} there would be strong selective pressure for a single display with different meanings among non-overlapping groups of recipients. Smith⁵⁻⁷ presents evidence that this is the case in many species of birds.

Morse^{1,8} states that song A occurs "chiefly in the presence of conspecific males and song B chiefly in the presence of conspecific females". But his data⁸ show that type B was given in the presence of females on only seven occasions while in "many" instances it was given by a male singing from an exposed perch (presumably in the absence of a female).

He also states¹ that song B increased in frequency in the dense population "at the peak of the incubation period, when there was little contact between sexes". In my observations of this species, I found that this song is usually given by males singing centrally on their territory, undisturbed (song B has been recorded in this context on forty-eight occasions, song A on sixteen occasions). Contrary to Morse's statement⁸, it is certainly given frequently by mated males; one male I observed in 1970 sang song B almost exclusively throughout the nest-building and incubation activities of his known mate.

My own observations support Morse's conclusion that type A song is given in the immediate presence of conspecific males. (Song A has been recorded in this context on fifty-eight occasions, song B never.) But it is also given in the absence of another male in certain other contexts: (1) when a male is singing alone at or near the boundary of his territory (song A on thirty occasions, song B on one), and (2) during the first bout of singing in the morning and the last bout in the evening (song A on thirty-one occasions and song B on three).

On the basis of these considerations and observations it is possible to give an alternative explanation of the causation of the two song types. I propose that a male black-throated green warbler, under the influence of a high androgen level, in the presence of adequate environmental conditions (the breeding habitat) and in the absence of conflicting external inputs, will spontaneously produce type B songs. But the additional presence of any of a number of external stimuli such as the presence of another male, low light level, or even being on "insecure" ground at the edge of the territory, will cause a switch to song type A or an increase in the proportion of type A songs delivered. The presence of another male may also produce an increase in the rate of song delivery. Intense territorial fighting, darkness, being off the territory or being non-territorial may cause cessation of singing. Absence of stimuli for type A singing will automatically result in the production of type B songs (during the singing period). The determination of the type of song may thus be seen as the outcome of the relative balance of two inputs; one internal (hormonal-neurological) and one external (sensory stimuli). My interpretation will explain the phenomena Morse observed without recourse to different ultimate functions for the song types. The high frequency of type A song in large populations compared with isolated males can be explained (as by Morse) by the sensory input (visual and auditory) from neighbouring males. The high absolute frequency of type B song in dense populations early in the season is explicable on the basis of a high internal hormonal input and a relatively lower external sensory input, the latter increasing as the breeding population becomes more dense (reaching a maximum at the end of May, when type B song reaches a minimum). Isolated birds are not as exposed to these external stimuli producing type A song as are birds in dense populations, and therefore sing relatively more type B songs early in the season.

The decline in type B songs in isolates during the season may be explained by a declining hormonal input.

The presence of other males is a general stimulus to sing, as is apparent from Morse's observation that the overall rate of singing is much higher in dense populations than in isolated

birds. The stimulating effect of song of one male on the singing of other males is well known both from natural observations and from play-back experiments^{9,10}. The increase in song B in dense populations after the first of June may result from the fixation of territorial boundaries and the resulting relative decline in situations of territorial interaction and situations where a male is on "uncertain" ground.

Observations of the contexts of the two song types, and particularly of the situations in which switching from one to the other occurs, suggest that the song types represent segments of a motivational continuum. This would suggest in turn that their function is rather similar. The function of territorial displays is to defend an area without resorting to physical encounters. The existence of more than one territorial display (such as different song types) allows the recipients to determine the activities and emotional state of the displaying males more precisely from a distance and to discriminate more finely in their responses. This would mean, for example, that there would be no reason to chase and fight to determine the position of a neighbouring male's territorial boundary.

At the same time there is nothing in such a situation to prevent both song types serving another function in mate attraction and pair-bond formation and maintenance, for the meanings of the displays depend on the classes of recipients.

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Phytogeographical Classification of Mesozoic Floras and their Bearing on Continental Drift

ALTHOUGH much palaeobotanical work has focused on Palaeozoic plant geography there have been few attempts to classify Mesozoic taphofloras. The Jurassic flora was thought to be uniform all over the world though Oswald Heer showed that the Siberian Jurassic vegetation (contrary to the contemporaneous European vegetation) had been dominated by the *Ginkgo* allies¹. Nathorst referred to the Mesozoic flora of Siberia and Arctic islands as "Phoenicopsis flora", and Prynada proposed the designation "Siberian" or "Phoenicopsis" flora for this type of Mesozoic vegetation. Vakhrameev^{2,3} coined the term "Indo-European floristic province" for the rest of Eurasian taphofloras of this age, uniting two floristic regions (India and Europe) of rather different aspect. I propose the following phytogeographical classification.

First, the Arctomesozoic or *Phoenicopsis* flora include the Siberian province of Prynada, Alaska, western Canada⁴ and

western Greenland⁵. One of the dominant genera of this flora was *Phoenicopsis*, a gymnospermous plant of Czekanowskian affinities. *Phoenicopsis* flora is supposed to be approximately equivalent to the temperate forest zone.

Second, there are the Madromesozoic or *Cycadeoidea* flora. The richest localities of *Cycadeoidea* and its allies (Monanthesia, but not the leptocaul bennettites which are widespread in all geofloras) are confined to the Black Hills of Dakota, the Freezeout Hills of Wyoming and north-western New Mexico^{6,7} not far from the area where Axelrod established his Madrotertiary flora⁸. The *Cycadeoidea* flora is well represented also in the European Jurassic and after considerable interval appears in India, Mongolia and Japan⁹. It is absent in the *Phoenicopsis* zone. The difference between these two zones seems to be vegetational as well as floristic. The Madromesozoic zone probably comprised the treeless areas of profuse growth of small pachycaul plants exposed to pronounced aridity, although most of the region was covered with conifer forests.

Third, there is the Notomesozoic or *Pentaxylon* flora. The *Pentaxylales* had been described from the Indian localities. They were identified in some southern hemisphere taphofloras, but never in the northern hemisphere apart from India. Their status in the southern floras was analogous to that of Czekanowskianes in the northern floras. Thus, the strong southern affinity of the Mesozoic Indian flora is evident.

The classification proposed here is relevant to the theory of continental drift. The fossil floras of peninsular India retained their southern affinity up to the Eocene¹⁰. The change of floristic composition of this area coincides with its supposed collision with Asia as the result of either anticlockwise rotation or the strike-slip along the transform faults¹¹. Thus, the palaeofloristic evidence agrees with the plate tectonic version of Indian plate history.



Fig. 1 Scheme of the Mesozoic geofloras.

Reservations must be made as to the other implications of continental drift. Some authors are inclined to explain all disjunct distributions, for example, distribution of the Proteaceae and other angiosperm groups, as the consequence of predrift (Triassic) arrangement of the southern continents¹². Such suggestions do not agree with the fossil record. No angiosperms were found in the southern taphofloras until the end of the Early Cretaceous. The similarities demonstrated between the Palaeozoic *Glossopteris* floras of Gondwana persist after the breakup in the post-drift late Mesozoic floras of this region. It seems that the floristic continuum was not greatly affected by the first stages of expansion due to successful migrations across the land bridges and/or "stepping stones". The Gondwana concept is not necessary for the explanation of all floristic connexions of the southern continents.

According to the plate tectonic theory, the lithosphere is consumed in the trenches bordering the Pacific Ocean, the consumption being superior to the crust accretion along the Pacific ridges. If the Atlantic Ocean is expanding, "it is axiomatic that another major ocean (Pacific) is contracting"¹³. If we accept this we might presume that the floras on opposite sides of the Pacific ocean have been more dissimilar in the Palaeozoic than they were in fact¹⁴. The *Cycadeoidea* flora is represented on both Asiatic and American margins of the

Pacific Ocean. There is a considerable gap in the fossil record of Cycadeoidea between Europe and Central Eastern Asia and the Cycadeoideas from Mongolia and Japan are probably of American origin. Migration across the Bering land bridge is excluded because the northern parts of Asia and America were occupied by the Arctomesozoic flora.

There is a striking parallel between the distribution of Cycadeoidea and horned dinosaurs, the latter being represented only in Mongolia and western North America¹⁵. According to Colbert, the horned dinosaurs crossed the Pacific Ocean but were incapable of surmounting the Lance Sea—a narrow strip of water. Migration along the Bering bridge is also doubtful because of climatic barrier.

These facts enable us to suggest the migration route across the Mesozoic Pacific Ocean at middle latitudes. This means a much narrower, not wider Pacific. It follows that the Pacific as well as Atlantic area was expanding at the end of Mesozoic time, the difference being probably not in the amount or crust accretion but in the arrangement of rise systems. While the true mid-ocean rise was active in the Atlantic Ocean, the asymmetrical seafloor spreading (half cell stile of spreading) took place along the margins of Pacific¹³.

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Revised Radiocarbon Chronology for the Stone Age in South Africa

SEVERAL radiocarbon dates have recently become available which necessitate a rather drastic revision of the chronology of the Stone Age south of the Limpopo River¹. An important consequence of the new time scale is that there is now strong evidence to suggest a very early appearance of *Homo sapiens sapiens* in Africa—considerably earlier than in the Middle East and Europe. It thus seems possible to envisage the continent of Africa as the area where much of modern man's early evolutionary development took place.

The first radiocarbon time scale for southern Africa was proposed in 1959 by J. D. Clark², who estimated that the Middle Stone Age (MSA) lasted from 38,000 bc to 6,000 bc, and that the Later Stone Age (LSA) postdated 6,000 bc. The only major subsequent modification of this scheme has been the shifting of the LSA/MSA interface back to about 10,000 bc³. When radiocarbon dates of greater than 50,000 years

were obtained for the MSA^{4,5}, however, the validity of the prevailing chronology was doubted, and these doubts have recently been confirmed by readings of earlier than 20,000 bc for LSA aggregates at three widely separated localities.

We have here, purely for present purposes, introduced various new terms, which are identified by means of quotation marks.

The LSA is usually defined in terms of the presence of flakes with plain striking platforms, partly produced by means of a punch technique⁶. The most consistent artefact classes are various convex scraper types, scaled pieces and bone points. Our analysis of the stratigraphic and dating evidence indicates that it is possible to subdivide the LSA into two major temporal phases, namely an "Early LSA" and a "Late LSA", equated typologically with the Upper Palaeolithic and Mesolithic of Western Europe, respectively.

The MSA, on the other hand, is distinguished by the occurrence of flakes with faceted butts, and the consistent, but not necessarily dominating, presence of flake points shaped either by convergent dorsal flaking or by lateral trimming⁶. Stratigraphic evidence provides proof that the MSA may be envisaged as an unbroken linear development, within which a number of temporal phases can be isolated on arbitrary typological grounds. It may in this way be separated into a "full MSA", representing the MSA proper, and a "final MSA", which would correspond with the now abandoned Second Intermediate.

The following sites have provided key dates for the "Early LSA" and the "final MSA" in South Africa:

(1) Rose Cottage Cave, near Ladybrand in the Orange Free State. At this locality three Wilton levels are successively underlain by a Pre-Wilton stratum, 1.5 m of beige sand containing for the most part a sparse and even older "Early LSA" aggregate and 3.0 m of "final MSA" ascribed to the Magosian^{7,8}. The pertinent dates^{4,5,9} are:

Pta-211	RC	1.35 m base Pre-Wilton	27,480 ± 520 bc
GrN-5300	RC	1.46 m top beige sand	23,690 ± 220 bc
Pta-354	RC	3.25 m base beige sand	> 39,000 bc
Pta-213	RC	3.33 m top Upper Magosian	> 48,250 bc
Pta-214	RC	3.78 m Upper Magosian	> 42,500 bc
SR-116	RC	3.78 m Upper Magosian	> 48,000 bc

These results show clearly that the "Early LSA" must have existed well before 27,500 bc.

(2) Heuningsneskrans Shelter, near Ohrigstad, in the Transvaal. Here 6.4 m of "Early LSA" deposit rest directly on bedrock and underlie surface Iron Age. The basal stratum yielded the following dates⁹:

Pta-114	Hnk.	1.56 m top Stratum 1	8,480 ± 150 bc
Pta-100	Hnk.	2.27 m upper Stratum 1	11,150 ± 110 bc
Pta-101	Hnk.	4.39 m middle Stratum 1	22,680 ± 300 bc

The apparently linear rate of accumulation, when extrapolated, gives a date of about 32,000 bc for the base of Stratum 1.

(3) Border Cave, near Ingwavuma, in Natal. The 1970/71 excavations at this site revealed an "Early LSA" white ash stratum up to 30 cm thick, lying between Iron Age and MSA layers towards the centre of the cave, and elsewhere a "final MSA" of Epi-Pietersburg type overlies the "full MSA"¹⁰. Dates for these levels are:

Pta-422 Ingw.	1.0 m top First White Ash	34,850 ± 1,000 bc
Pta-433 Ingw.	1.1 m middle First White Ash	34,150 ± 900 bc
Pta-424 Ingw.	1.2 m base First White Ash	33,750 ± 1,100 bc
Pta-459 Ingw.	0.4 m base Epi-Pietersburg	> 46,300 bc

(4) Montagu Cave, near Montagu, in the Cape. Here Wilton (Layer 1) is underlain in turn by up to 1.5 m of grey sand containing "final MSA" of Howieson's Poort aspect (Layer 2), and by two Acheulian levels separated by sterile red sand (Layers 3–5). Layer 2 was subdivided into an upper part, with seven surfaces represented by increased artefact

concentrations, and an undifferentiated lower portion^{11,12}. The apposite dates are^{4,12,13}:

GrN-4726 MC	1.55–2.24 m	Surface 1, Layer 2	21,250 ± 180 BC
GrN-5123 MC	1.77 m	below Surface 6, Layer 2	17,150 ± 110 BC
GrN-5124 MC	1.78 m	below Surface 7, Layer 2	> 48,850 BC
GrN-4728 MC	3.1 m	base, Layer 2	43,950 ± 2100 BC
GX-0947 MC		base, Layer 2	> 36,050 BC

The second date (GrN-5123) is clearly anomalous and should be disregarded. The three samples from the lower part of Layer 2 could not have been derived from the underlying Layer 3, because this contains no organic material¹². The possibility of admixture of younger charcoal from Layer 1 with the two upper samples cannot, however, be excluded, especially as the surfaces slope sharply. The results prove that at least the beginnings of the "final MSA" at this site antedate 40,000 BC.

(5) Klasies River Mouth Cave, near Cape St Francis, in the Cape. The broad cultural sequence at this site is "full MSA" (Mossel Bay), then "final MSA" (Howieson's Poort), and finally LSA¹⁴. An unpublished series of dates, all around 36,000 BP, has been reported for the Howieson's Poort level¹².

(6) Howieson's Poort Shelter, near Grahamstown, in the Cape. At this type locality about 25 cm of surface sand overlies 15 cm of grey earth containing Howieson's Poort¹⁵. A single date is available¹⁶:

I-1844 HP 38 cm Layer 4 16,790 ± 320 BC

This reading would seem to contradict the other dates for the "final MSA", but the strata are thin and it is possible that the dated material derived partly from the overlying level, which was reportedly rich in organic debris.

The available data do not enable the LSA/MSA interface to be defined accurately, but the evidence supports a dating of before 35,000 BC. The artefact classes from the basal levels of all three "Early LSA" sites show no marked affinities with "final MSA" types. From this may be deduced, either that the transition between these two traditions took place at an even earlier date or that an already developed "Early LSA" intruded into South Africa shortly before 35,000 BC.

On the basis of the new dates it seems likely that the "full MSA" falls entirely beyond the effective range of the radiocarbon method—a finding in accord with most of the available readings for this phase, bearing in mind that all measurements in the 40,000 yr range should be regarded as minimum dates only. Rough estimates, based on apparent sedimentation rates at certain sites^{4,9}, and associations with raised beaches⁶, suggest that the onset of the MSA may well date back to beyond 100,000 BC.

The MSA is distinguished by dynamic typological innovation. Occurrences ascribable to its beginnings have affinities with some Middle Palaeolithic industries, but subsequent phases reveal the introduction and development of flaking techniques and tool forms which foreshadow the LSA and Upper Palaeolithic, such as punch-struck blades, backed knives of Abri Audi and Chatelperronian types, burins and scaled pieces.

Of considerable interest is the association of *Homo sapiens sapiens* remains with the MSA. The Florisbad skull probably derives from Peat Bed 1, which yielded an aggregate best ascribed to the very onset of the MSA, dated to more than 46,950 BC (GrN-4208)^{4,17}. At Tuinplaats "advanced MSA" artefacts were found at levels higher than the remains of a robust individual¹⁸. Border Cave has yielded a partial adult cranium possibly belonging to the MSA, as also an infant skeleton from a deliberate burial, dating either to the very base of the Epi-Pietersburg or to the underlying "normal Pietersburg"¹⁰, and therefore older than 46,000 BC. The MSA deposits at Klasies River Mouth Cave have produced two mandibles which apparently predate 34,000 BC^{12,19}. An almost complete skeleton was found in the Howieson's Poort level at Skildergat Cave^{6,20}.

That comparable forms are not confined to South Africa is indicated by the remains recovered from the Kibish Formation in the Omo Valley of Abyssinia. These have been dated to older than 33,000 BC (L.1203A)^{21,22}.

It therefore seems likely that *Homo sapiens sapiens* already occupied much of Africa by the advent of the "final MSA", that is, before 50,000 BC at the latest, and there are indications that his origins may even reach back to the beginnings of the "full MSA", at probably more than 100,000 BC. This opens up a new perspective in palaeoanthropology, because nothing of comparable age which can be related to modern man has so far been found in Europe or South-West Asia, with the exception of the apparently transient intrusion of the Amudian (Pre-Aurignacian), as at Jabrud and et-Tabun, in Syria. The radiocarbon evidence places the advent of the Upper Palaeolithic at 33,000 BC in West Europe, at about 38,000 BC in the Middle East and at perhaps 42,000 BC in central Europe²³.

It is concluded that the new radiocarbon dates discussed here indicate: (1) that the onset of the Later Stone Age in South Africa predates 35,000 BC; and (2) that the typological transformation represented by the Middle Stone Age and the physical evolution of a basic grade of *Homo sapiens sapiens* were concomitant events which took place in Africa over a protracted but as yet undefined period before 35,000 BC.

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Uranium-series Dating of Bone from the Isimila Prehistoric Site, Tanzania

EXCAVATIONS in 1957 and 1958 at the Isimila prehistoric site, in Tanzania¹, sampled Acheulian occurrences in horizons at various levels in the Isimila Beds which are approximately 18 m

thick. No significant breaks were observed in the sedimentary sequence, although there are numerous local hiatuses.

The aggregates of artefacts from the upper beds (Sands 1) were broadly correlated geologically and typologically with the upper Acheulian aggregates from the Kalambo Falls prehistoric site, where minimum ^{14}C dates of $61,700 \pm 1,300$ [GrN 4896] and $60,300 \pm 750$ yr [GrN 2644] have now been obtained^{2,3}, and with the upper archaeological occurrences at Olorgesailie. All of these were compared with "stage 9" from Bed IV at Olduvai Gorge⁴. Considerable work has been done at these sites since 1958, including further work in Sands 3 at Isimila⁵. The work at Olduvai is of particular relevance to the original dating suggested for the occupation sites in eastern Africa, because current work in Bed IV by M. D. Leakey and in Bed III by Kleindienst⁶ indicates that the "stages" of the Acheulian which are recognized at Olduvai Gorge are invalid on stratigraphic grounds. The time of deposition of Sands 1 at Isimila may be considerably earlier than 60,000 yr BP.

Although we recognized at Isimila that the relatively small samples of artefacts obtained from archaeological occurrences in Sands 2, 3, and 4 differed typologically from those in Sands 1, the absence of any marked unconformity or developed weathering horizon separating Sands 1 and the horizons below suggested that the Isimila Beds were deposited over a short time span. The subsequent discovery of artefacts at Isimila and at the other sites favours a greater time span; we did not envisage quite as old a date for Sands 4 as that obtained by the uranium-series method but, insofar as the archaeology is concerned, have no reasons for rejecting it.

The dated sample, bone fragment from the upper half of Sands 4, was exposed in Trench I in grid unit G. 23-3. Bone is only infrequently preserved at this locality, and almost all of the few specimens recovered by us were from Sands 4. Some preliminary identifications have been published¹; they include specimens of a proboscidean, hippopotamus, a suid, an equid and bovid(s). The samples have been submitted elsewhere for further study and identification. Such work could also be very useful in assessing the relative biostratigraphic age of the mammalian assemblage represented, and hence be of some relevance in interpreting the results of the radiometric age determination. One of us (K. P. O.) was asked by Professor F. Clark Howell to undertake analytical dating of the fossil bone samples. Radiometric assays were carried out in the Anthropology Sub-department of the British Museum (Natural History) and fluorine and phosphate analyses were undertaken in the Laboratory of the Government Chemist.

Table 1 Analytical Results on Fossil Bones from Sands 4 at Isimila

Sample	F %	$100 \times \frac{F}{P_2O_5}$	eU_3O_8 * p.p.m.
T2 H20-3	2.6	7.5	351 ± 4
T6 H20-3	3.0	7.8	485 ± 5
G23-3	3.0	8.2	$1,149 \pm 6$

* Counts of beta radiations were calculated as equivalent uranium oxide in parts per million.

The fluorine content of the samples is rather similar, which is consistent with their all being of the same stratigraphical age (see Table 1). The uranium content varies considerably, which indicates that these bones were exposed to different dosages of uranium-family elements through varying local conditions.

At the request of the British Museum (Natural History), one of us (B. J. S.) undertook dating of bone sample G23-3 by the uranium-series method. Dating fossil bones by this method has been tried earlier⁷⁻⁹ assuming an ideal closed system with respect to uranium and daughter elements. But it was reported recently¹⁰⁻¹² that some of the dated bones showed evidence of uranium migration; thus the measurements of both $^{230}\text{Th}/^{234}\text{U}$ and $^{231}\text{Pa}/^{235}\text{U}$ activity ratios were required for reliable dating.

The analytical procedure used for this dating has been described in detail previously^{10,13}. Table 2 shows the results,

Table 2 Analytical Results and Calculated ^{230}Th and ^{231}Pa Ages of Bone Sample G23-3

Uranium p.p.m.	Thorium p.p.m.	$^{234}\text{U}/^{238}\text{U}$	$^{230}\text{Th}/^{234}\text{U}$	$^{230}\text{Th}/^{232}\text{Th}$	$^{231}\text{Pa}/^{235}\text{U}$
$1,457 \pm 20$	0.248 ± 0.012	1.23 ± 0.02	0.96 ± 0.05	21,000	1.01 ± 0.04

Calculated ^{230}Th age: $260,000^{+70,000}_{-40,000}$ yr.

Calculated ^{231}Pa age: older than 170,000 yr.

together with the calculated ^{230}Th and ^{231}Pa ages. The uranium concentration is rather high, 1,460 p.p.m. The common thorium, ^{232}Th , concentration is low, 0.25 p.p.m., which indicates that essentially all ^{230}Th was produced by the radioactive decay of ^{234}U and ^{238}U isotopes in the sample. The $^{231}\text{Pa}/^{235}\text{U}$ activity ratio is unity, within limits of error, indicating that the specimen was undisturbed during at least the past 170,000 yr. The ^{230}Th age of the bone is calculated from the measured activity ratio of $^{230}\text{Th}/^{234}\text{U}$ to be $260,000^{+70,000}_{-40,000}$ yr old.

The initial radiometric assays on Isimila bone were conducted in the British Museum (Natural History) with the aid of a grant from the Wenner-Gren Foundation, New York.

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BOOK REVIEWS

A Critical Problem

Man's Impact on the Climate. Edited by William H. Matthews, William W. Kellogg and G. D. Robinson. Pp. xvii + 594. (MIT: Cambridge, Massachusetts and London, December 1971.) £9.

THE widespread growth of public concern over what man is doing to his environment, partly under the pressures of the population explosion but so far perhaps mostly through the habits of an economy which demands continual industrial expansion for its healthy working, must now include an interest in whether the world's climate may also be affected. As it is, most planners are probably aware that there have been changes of climate, whether natural or caused by human activity, in this century, which have changed the geographical limits of species in the natural flora and fauna, and the economically determined limits of certain crops, and which have been accompanied by a rise of the global sea level that has been an increasing threat to low-lying lands (and cities) and raises anxious problems regarding coast defences.

The Massachusetts Institute of Technology has been notably active in sponsoring working conferences to consider these matters—a Study of Critical Environmental Problems (SCEP) held at Williams College, Williamstown, Massachusetts, in July 1970, and a Study of Man's Impact on Climate (SMIC) at Wijk near Stockholm in July 1971—followed by presentation of the results published in book form^{1,2} (by the MIT Press) with astonishing rapidity afterwards. The work here under review incorporates the report and recommendations of SCEP and adds a series of papers presenting more detailed information on the many diverse aspects of the problems discussed at the two conferences. There are forty-eight chapters in all by different authors and varying in length and depth, partly as dictated by the present state of knowledge in the fields concerned. There is a companion volume, *Man's Impact on Terrestrial and Oceanic Ecosystems*, which similarly reproduces and supplements those aspects of the SCEP report concerned with the more direct impact of human activity on the biosphere. All these works represent a substantial and timely effort to provide, in brief summary form, the essential information

(derived from studies in many disciplines) to guide not only researchers and students but practical men, scientists and others, who face the political responsibility to take the decisions that may be necessary to avert possible disasters.

The thirty-nine contributors to the volume here reviewed, many of them chief scientists in various American laboratories (but including one in Sweden and one in Germany), include about a dozen of the leading names in meteorology and others equally eminent in radiation, atmospheric chemistry and the problems of particulate pollution. After some introductory chapters containing material from the SCEP report, Part II deals with the factors that determine climate change and how man might affect them. Parts III and IV are devoted to the contributions to our understanding that may be derived from mathematical modelling and monitoring of the atmosphere's behaviour respectively. There follow chapters devoted to information on specific pollutants that may affect climate. A final section is interestingly turned towards the difficult social and political issues which scientists and other types of citizen in positions of responsibility must come to understand and face together.

Throughout the book the style is commendably lucid and direct. The carefully selected diagrams are mostly simple to understand and highly informative, though a number of them are inevitably only sample surveys, individual case studies or first attempts at world surveys from all too few measuring stations operating over only recent years. Inadequacies of knowledge are made clear. One must feel uneasy about the state of our interpretation of the atmosphere's heat budget and general circulation, when the best observation-based estimates of the Earth's over-all albedo have been changed over a few years from about 43 per cent to 29 per cent (especially when it is thought that only about a 2 per cent change in the solar radiation available would suffice to cause glaciation of the whole Earth).

A couple of quotations from the chapter by J. Murray Mitchell on climatic changes and causes of change will show the quality of the book and its wisdom in applying the present state of knowledge to the problems in hand. Mitchell first indicates the often

neglected point that we cannot hope to isolate man-made changes until we can trace the background of natural fluctuations of climate that are forever going on. "The changes of climate from one geological epoch to another, and apparently also those from one millennium to another, are clearly too large to be explained as random excursions from modern norms . . . one might be led to suppose that . . . changes of climate associated with the alternating glacials and interglacials . . . are smoothly varying functions of time, readily distinguishable from the much more rapid variability of year-to-year changes. . . . In such a case the averaging interval needed to obtain a stable estimate of present-day climate should be long enough to suppress year-to-year sampling variability, but short in comparison to . . . a glacial cycle. If we succumbed to the foregoing rationale, we should probably be living in a fool's paradise . . . the apparent regularity of atmospheric changes in the geological past is only an illusion, attributable to the inadequate resolving power of palaeoclimatic indicators. . . . The atmospheric state is known to vary on many scales of time." The writer goes on to distinguish two views of climatic change: (1) the "slave" concept, whereby there is only one equilibrium state for the atmosphere under a given set of Earth-environment conditions, and (2) the "conspirator" concept, suggested by E. N. Lorenz (another contributor), under which alternative equilibrium modes exist for the atmosphere under one and the same set of environmental conditions and the outcome, as to which mode prevails, depends on the antecedent history including possibly random transient environmental disturbances and/or excursions of the atmospheric state along the way. The latter possibilities might well, and presumably would, prevent any confident diagnosis of cause and effect in the case of observed climatic variations, set up either by natural environment changes or the actions of human beings. Mitchell suggests that, because there is no real possibility of detecting Lorenz-type transient disturbances, we must be bold enough to assume they do not occur and to use the available estimates of the specific climatic effects of this or that identifiable environmental change whether natural (for example, volcanic) or man-made (for example,

this or that pollutant). We should use these considerations to alert ourselves to climatic dangers which might otherwise be totally unsuspected (though we may retain a reasonable scepticism about them) and to guide our efforts at monitoring what happens. The latter should provide the firmer knowledge which decisions on action demand.

H. H. LAMB

¹ *Man's Impact on the Global Environment: Assessment and Recommendations for Action (Report of the Study of Critical Environment Problems (SCEP))* Library of Congress catalog card number: 74-139447 (MIT Press, Cambridge, Massachusetts, 1970).

² *Inadvertent Climate Modification (Report of the Study of Man's Impact on Climate (SMIC))*. Library of Congress catalog, card number: 79-170861 (MIT Press, Cambridge, Massachusetts, 1971).

Lignin Chemistry

Lignins: Occurrence, Formation, Structure and Reactions. Edited by K. V. Sarkanen and C. H. Ludwig. Pp. xiv + 916. (Wiley: New York and London, September, 1971.) £16.50.

EDITORS Sarkanen and Ludwig are to be commended for undertaking and completing successfully the task of drawing on the resources of nineteen authors to produce an up-to-date definitive picture of the occurrence, structure, reactions and utilization of lignins. No one with an interest in the many and varied aspects of the plant kingdom can ignore the significant role of the lignin component. The botanist, the biologist, the plant anatomist, the plant physiologist, the microbiologist, the biochemist, the chemist, the engineer and the industrialist will each find significant material of interest to him in one or more of the twenty chapters of this book. Despite this broad coverage, any reader with a desire to learn more about the present status of the many areas of study involving this abundant natural resource, is aided by the similar writing style used throughout by the nineteen contributing authors. Occasional, but useful, cross-referencing between chapters may also be found. Needless to say, the book will be used primarily as a reference volume, to which each individual will turn for his own particular need.

For me, the contribution of this book is that each chapter has been written with a recognition and appreciation of the value of modern methods of study of molecular structure and has thus made clear the rightful place of lignins as interesting natural polymers. For those who in the past may have lost interest and become disillusioned in their study of lignins, this book, if read carefully, should rekindle the

spark for further and enthusiastic research.

The nineteen authors, each well recognized for his particular interest in the lignin problem, have critically reviewed the massive lignin literature and presented a well referenced and useful analysis of the present state of knowledge. The book is divided into nine parts beginning with a discussion of the problems involved in arriving at acceptable definitions and nomenclature for lignin and related compounds. For the first time this difficult problem has been critically examined and some useful suggestions made for its rationalization. In part two, chapters on the occurrence and formation as well as classification and distribution of lignins, provide the extensive background for initial studies in this field. Current theories of lignin biosynthesis and methods of isolation are discussed in part three.

The section on spectroscopic characterization is particularly valuable in the light of the rapidly increasing application of these techniques to structural studies. Unfortunately, no reference is made to the more recent but potentially valuable use of mass spectrometry especially with reference to the identification of lignin degradation products.

Reviews in part five on solvolysis, halogenation and nitration, oxidation, reduction and hydrogenolysis, modification reactions and high energy degradations serve to up-date similar discussions found in earlier monographs. Part six is directed to those more interested in reactions involved in commercial pulping processes both sulphite and alkali, but with extensive attention given to the probable mechanisms involved and the nature of the resultant products. The polymer properties of lignin and lignin derivatives are the subject of part seven, and, in a macromolecular sense, relate the application of interesting physical chemical techniques to theories regarding nature of the lignin in the wood. The fate of lignin resulting from microbiological attack is discussed in part eight in the biological transformation of lignin. The last two chapters, comprising part nine, will be of particular interest to the industrial chemist and chemical engineer since they deal with the utilization of lignin, both as a useful polymer and as a source of low molecular weight chemicals.

It may be argued that this book contains much of the material to be found in other earlier monographs. To a degree this is true, but there is significant updating of the published data and, more important, a real attempt made throughout to emphasize the scientific and economic rewards to be gained by continued research in this field.

Relatively few and minor errors have

been noted in the book, none of which should lead to any misinterpretation of meaning. I feel, however, that the manner in which the several tables and figures are reproduced is unfortunate. The poor quality of these compared with the clear, easily readable type of the rest of the material is very noticeable. For such an important compilation, which will undoubtedly become the prime source of reference for lignin scientists for years to come, one would have hoped for a better overall impression.

The editors have chosen to honour Professor Karl Freudenberg by means of a full-page photograph, rightfully acknowledging his "unparalleled series of contributions" to lignin research.

J. M. PEPPER

Chloroplasts

Structure and Function of Chloroplasts. Edited by Martin Gibbs. Pp. xv + 286. (Springer: Berlin and New York, 1971.) 80 DM; \$24.40.

If the distinguished editor of a distinguished journal is invited to edit a book relating to his own speciality, it is not too much to hope that it will be first class. In fact *Structure and Function of Chloroplasts* edited by Martin Gibbs is impeccable.

In the late 1950s it must have seemed to some that, with the discovery of photosynthetic phosphorylation and the elucidation of the reductive pentose pathway of carbon assimilation, the principal problems in photosynthesis were solved. Instead, as so often happens, this important and fundamental work was to provide the foundation for a great many new advances. Emerging from these now is a growing preoccupation, not only with the multiplicity of biosynthetic reactions and ion movements within the chloroplast but also with the manner in which such processes affect the chloroplast relationships with its immediate cytoplasmic environment. It is with these aspects that the book is concerned, rather than those photochemical events which are completed within fractions of a second of the light excitation of a chlorophyll molecule.

In characteristic individual style, Robert Hill introduces the collected papers with a historical outline which, in six brief pages, still manages to range from the fundamental contributions of workers like Robert Emerson to the eccentricities of Nathan Pringsheim who once suggested that "chlorophyll by absorbing light protected what might be a colourless photosynthetic system". This introduction is embellished (at what cost in time and effort to its author?) by the full given names of each contributor listed.

Kurt Mühlethaler then summarizes present interpretations of the "Ultra-structure of Plastids" (including proplastids, amyloplasts, proteinoplasts, chromoplasts and so on, as well as chloroplasts) providing a very useful picture of the structural framework to which the remaining articles may be related. This is followed by Frank Mayers's account of "Light-Induced Chloroplast Contraction and Movement", a welcome inclusion, containing a surprising wealth of information, on a topic which has defied all but the most stoic investigators since the first observations in 1850.

Two articles, one by Björn Walles on "Plastid Inheritance and Mutations", the other by Christopher Woodcock and Lawrence Bogorad on "Nucleic Acids and Information Processing in Chloroplasts", are so comprehensive and useful that they will undoubtedly sell this volume in their own right to workers in these areas. Similarly there are few investigators in my own general field who could resist the appropriate juxtaposition of Mordhay Avron on "The Biochemistry of Photophosphorylation" and Martin Gibbs on "Carbohydrate Metabolism by Chloroplasts". The article by Avron is excellent but regretably short. Avron is a leading exponent of the view that experiment is better than conjecture and, at this stage in the elucidation of the relationship between electron transport and phosphorylation, this view can only be endorsed. Opinions based on Avron's formidable experimental experience, however, are always invaluable and on this occasion his otherwise admirable reticence is the reader's loss. The article by Gibbs covers the ground with customary authority and includes valuable comments on the present convolutions of "C4 photosynthesis" and also on the transport of metabolites between chloroplast and cell.

In recent years Andrew Benson's name (formerly linked with that of Calvin and the path of carbon) has become increasingly associated with chloroplast lipids and his chapter on this subject is consistent with his high standing in this field.

Trevor Goodwin concludes the book with a *tour de force* on "Biosynthesis by Chloroplasts". This includes sections on protein synthesis and lipids which complement those by Walles, Woodcock and Bogorad and Benson and leads naturally to the chloroplast pigments and to the study of terpenoids to which Goodwin has contributed so much.

Within the limits which it defines this book achieves virtually all that it sets out to do. One of its strengths is that it will not only give the specialist articles of the highest standard but will also provide, between the same covers, the

immediately relevant background information which will allow him to start to build a picture of the chloroplast as a whole. D. A. WALKER

Diversity of Plants

Plant Speciation. By Verne Grant. Pp. x+435. (Columbia University: New York and London, August 1971.) £7.25.

UNTIL comparatively recently, there was a tendency to divorce discussions of the phenotypic course of evolution from considerations of its underlying mechanisms. The first was largely the province of the taxonomist and palaeontologist while the second received attention from more genetically inclined investigators of two kinds—those principally concerned with the consequences of selection and those more interested in the processes of heredity. The first of these genetical approaches, frequently adopted by zoologists, was characterized by considerations of gene frequencies, major gene polymorphisms and, at least, quadratic equations. The second attitude, more often favoured by plant geneticists, was expressed in discussions of breeding systems and chromosome mechanisms.

The content and approach of this book can be viewed in these terms. But the author's previous experience as a geneticist at the Rancho Ana Botanic Garden and Director of the Boyce Thompson Arboretum puts him in an admirable position to combine the attitudes of taxonomy and genetics. Further, his wide experience and detailed investigations of *Gilia* contribute considerably to the qualities seen in the present book which is intended to be complementary to his earlier work, *The Origin of Adaptation*.

There is little in *Plant Speciation* of which one can complain but it includes the use of the word "clone" to describe the products of uniparental reproduction whether these are produced asexually or by self-fertilization and the mistaken view that intergenomic pairing (that is allosyndesis, in my opinion) is synonymous with heterogenetic pairing. By inference the author would presumably equate intragenomic pairing (that is autosyndesis) with homogenetic pairing which is clearly a nonsense. The one set of terms refers to chromosome homology (or homoeology), the other to genic relations.

It is also unfortunate that the term incompatibility is used in relation to both crossability barriers between mating groups and mechanisms which impede or prevent inbreeding (for example, self-incompatibility) within them. Clearly, these two phenomena are quite distinct; members of separate mating groups are isolated by their

differences but members of the same group are prohibited from direct gene exchange by their similarities.

There is also some tendency towards inconsistency between various ideas expressed in the book. For example, it is claimed (page 121) that the two forms of sympatric genetic differentiation (intra-specific polymorphism and species differences) differ qualitatively in that the former is "segregational variation" while the latter involves "complex gene combinations". Yet elsewhere (for example, on pages 65 and 175) the author stresses that "species often differ allelically with respect to linked morphological and viability genes" and attention is expectedly paid to semi-species. Admittedly, there are clear examples of qualitative differences between the two variation patterns, for a sexual dimorphism can no more constitute a species difference than a polyploid series can compose an intraspecific polymorphism—except as a taxonomic artefact. But the significant difference between the two patterns of sympatric variation is neither quantitative nor qualitative but functional (compare incompatibility above). Differences between morphs, by creating a mutual dependence, are a means of maintaining the integrity of a mating group while those between species create competition and interrupt the genetic continuum. The inevitable qualitative difference between the two states is that the former unavoidably involves a balance between often-favoured heterozygotes and one or more homozygotes while the latter generally consists of two or more alternative homozygous states at the (chromosome level) with selections acting against the hybrids and/or the means of producing them.

The standard of illustration is high but there are disappointingly few tables. The general treatment, however, is well balanced, the information is presented in an orderly manner and the material is generously discussed. The book, in fact, represents a useful contribution in an area of enquiry shared by population geneticists, taxonomists and natural historians. K. R. LEWIS

Biological Cybernetics

Information and Control in the Living Organism. By Bernhard Hassenstein. Pp. viii+159. (Chapman and Hall: London, December 1971.) £1.30.

THIS elementary introduction to biological cybernetics by the well-known zoologist of Freiburg University might well be the best way to arouse the interest of young biologists in general cybernetics. Books on this subject are often in forbidding mathematical language

age and dominated by electronics and communication engineers who do not know the far from exhausted possibilities of control and information theory at many levels of life's organization.

A distinguishing feature of the book is that such basic concepts as signal transmission, feedback control, information and its storage, noise, redundancy, data process and many others are mostly introduced using examples either from sensory-motor skills or from language, that is from general or easily acquired experience. The emphasis on the biological aspect of cybernetics has led the author to develop graphic symbols, which in some ways differ from those usually employed in technology.

Some useful cybernetic concepts such as "effERENCE copy" or "learning matrix", which are current in the German literature, may be new even to the advanced reader only familiar with English language publications. The biological systems in which cybernetic principles have been found to be most conspicuously operative naturally provide most of the examples illustrating them; they are on the level of functional organism (the neuromuscular system, the brain, the endocrine glands) and on the developmental level (the genetic code). Social and economic applications of cybernetic concepts are mentioned, but not further pursued.

The last pages of the book are particularly useful for the novice in the subject, which is a heady one and has in the past generated more heat than light. Hassenstein emphasizes the importance of a neutral terminology for cybernetic deliberations, rather than the mere transfer of special concepts developed in one field to another. In his opinion it is not useful to speak of "electronic brains", "artificial intelligence" or "thinking machines". The words brain, intelligence, thinking, pertain to human psychology and cannot simply be transferred to machines. In neutral terms it can, however, be stated that these three devices can, like the human brain, process information.

This book is well translated and reads easily. There is, however, one slight ambiguity on p. 73, where the word "letters" could at first glance mean sheets of paper and not symbols. There are many line drawings, a good subject index and a useful list of more advanced books and papers in biological cybernetics.

H. KALMUS

Plain Man's Geophysics

Restless Earth: a Report on the New Geology. By Nigel Calder. Pp. 152. (British Broadcasting Corporation: London, February 1972.) £1.90.

NIGEL CALDER's book is claimed to be

written in plain language in order to bring reports to the layman from the very forefront of scientific research. He certainly does this and he does it with the verve, enthusiasm and understanding which have characterized his previous books. The book is lavishly illustrated with colour, black-and-white plates and numerous diagrams which, with virtually no exceptions, add both to the appearance and comprehension of the topics which are discussed. These topics are vast and numerous, concentrating on the acceptance of the new version of continental drift, that is, plate tectonics, but using this to range over most aspects of the Earth from its origin to man's present depredation of its resources. In this way, Nigel Calder puts an essential background to the present polarization of views on pollution, conservation, and so on.

As a book for the non-scientific layman, the praise must be fulsome and any adverse criticism arises mainly from the standpoint of the somewhat more scientifically minded layman or school reader, for which it is not really satisfactory. This is because a conventional journalistic technique has been used in which the theme jumps discontinuously from topic to topic, each section being in itself logical, but there being no coherent pattern running throughout the book or even in individual chapters. To take only one example, earthquakes are discussed on every tenth page, or so it seems. This discontinuous style gives a liveliness to the text and the reader certainly gets the feeling that everything is on the move (as it is!) but if "man is by nature a scientific animal" (p. 44), then a more logical development would have been more suitable. There is an understandable overplaying of the newness of the concept of plate tectonics. After all, geologists (people "studying facts in search of a theory") were only pushed against continental drift by geophysicists who now demonstrate its reality. This human interplay is sadly neglected—but does this really matter for the non-scientific layman? Perhaps it does to the extent that it would have led Nigel Calder to be much less dogmatic about our present day knowledge—is it really true that there is no longer uncertainty about the origin of earthquakes and volcanoes (p. 17) and can high mountains only be created by horizontal movements (p. 23)? Many ideas which are, at best, hypotheses are presented as facts and surely a healthy degree of scepticism should be presented to all levels of readership? A more continuous, logical theme would reduce some factual errors, but the number of these are incredibly few and the claim for up-to-dateness is clearly justified when several aspects have yet to appear in academic literature. For the non-scientific layman, the book is most

highly recommended and the BBC must be congratulated on an excellent production; it is a pity that it cannot be so acclaimed for a higher level of readership.

D. H. TARLING

Insect Development

International Journal of Insect Morphology and Embryology. Edited by A. P. Gupta. Pp. 97. Vol. 1. No. 1. (Pergamon: New York and Oxford, 1971.)

BIOLOGISTS do not find themselves short of mediocre reading material. Those planning a new journal, such as *Intern. J. Insect Morph. Embryol.*, should therefore examine their motives critically. In the Editor-in-Chief's words this new journal "marks the fulfilment of a dream which took over a decade to materialize". Can we share his enthusiasm? It seems reasonable to ask that any new journal should either provide a forum for a new field or raise the standard where quality is lacking. Haven't we enough journals covering embryology and morphology? Do we want to provide a receptacle for those insect papers that other biologists might read if they were in a general journal but certainly won't read if they are in a specifically entomological journal?

If we examine the contents of the first issue it may help us assess the benefit to biology of this new journal. There are two papers on ultrastructure, one on sperm maturation and one on nerve: both would receive a more general readership if published in an ultrastructural journal. There are three papers dealing with the structure of the insect gut and a couple on comparative morphology of two organs that are only found in insects. It is true that these latter five papers are only likely to be of interest to entomologists, but many descriptive papers are only of interest to specialists, and this fact does not constitute an argument for devoting a morphological journal to every large taxonomical group. These papers might find a home in the *Journal of Morphology*, for example.

Even in the first issue the standard of the papers, and their presentation, is mixed so that the journal seems unlikely to justify itself on grounds of excellence. One example: in a series of poor half-tones depicting scruffy sections through a wasp's head, it is hard to distinguish the pulverized bits of wasp from the dust on the microscope.

I think we can do without this journal, and it would therefore be better if we did. It remains of course to be seen whether authors flock to publish in it, but I hope the page charges put them off.

P. A. LAWRENCE

CORRESPONDENCE

Dating Error

SIR,—In the letter "Fission Track Dating of Archaeological Materials from Japan" (*Nature*, **230**, 242; 1971), Nishimura states that "... several questions still remain unsolved". Indeed, several points remain unresolved to these readers.

Following the author's method of age calculation, the data in Table 1 for the glass ball, Hirono, Uji, Kyoto Pref., indicate an age of 180,000 years, not 1,480 years. Also, for the glaze on bowl, Isumi, Toki City, the data indicate an age of 47,000 years, not 400 years. In addition, there seems to have been an interesting but unknown factor used when rounding-off figures for several of the other ages.

When calculations of uranium concentrations are made from the data in Table 1, uranium concentrations of up to 0.7% in several slags and up to 10% in zircons were found. These values are unusually high and subject to suspicion.

Although the author did not specifically mention the method of counting, one can assume from the reported etching conditions that an optical microscope was used. To our knowledge and experience, it is impossible to resolve accurately and record fission tracks with densities of 10 million to nearly 1,000 million tracks cm^{-2} , using an optical microscope.

Finally, we would like to emphasize that fission track dating of any material is possible only when $\text{Cu} \cdot T > 10$, where Cu =uranium concentration by weight % and T =age in years (for minimum track density of ten tracks cm^{-2}). That is, in order to date a material 1,000 years old, the uranium content of the material must exceed 100 p.p.m.

Yours faithfully,

GUNTHER A. WAGNER

MICHAEL REIMER

Department of Geology,
University of Pennsylvania,
Philadelphia 19104

Dr Nishimura replies:

SIR,—I apologize for my mistakes, and comment as follows.

Application of the fission track method to archaeological remains:

(i) For the baked relics, zircon was separated from the baked relics using the standard separation methods: heavy liquids and isodynamic separator. A few hundred grains of zircon were collected from a sample. A part of these minerals (usually about ten to twenty grains) was irradiated by neutron flux, and etched by concentrated P_2O_5 at $450^\circ\text{--}480^\circ\text{C}$ for the determination of suitable etching time. The other portion was etched on the basis of this test. The minerals containing one or more spontaneous fission tracks were selected under the microscope. The induced tracks in these minerals were counted after neutron bombardment.

(ii) For the glass, the glasses were mounted in a clear epoxy resin on a polymer slide and the mount was then ground and polished to expose the interior of the glass. A small part of this glass was irradiated by neutron

flux and was etched by 46% HF at 25°C for the determination of suitable etching time. The other portion was etched according to the results of this test. After the sample had been etched and washed, it was ready for counting. This procedure was repeated several times to obtain suitable counts. The induced tracks in the sample were counted after neutron bombardment and polishing.

For counting tracks, a differential interference microscope with mesh micrometer was used after, and I made errors in calculation of the counting area. I appreciate Wagner's and Reimer's pointing out my mistakes and present a corrected Table 1 for the letter to *Nature* (**230**, 242; 1971).

Yours faithfully,

S. NISHIMURA

Institute of Earth Science,
Kyoto University,
Kyoto

Table 1 Fission Track Ages of Archaeological Materials from Japan

Sample	Locality	ρ_s (cm^{-2})	ρ_t (cm^{-2})	ϕ (cm^{-2})	Fission track age (yr BP)
Zircon (baked earth)	Ise, Hata, Mie Pref.	4.7×10^3	5.7×10^7	0.46×10^{15}	2300
Zircon (baked earth)	Hirakata, Osaka Pref.	2.0×10^3	2.5×10^7	0.46×10^{15}	2200
Zircon (baked earth)	Hisai, Mie Pref.	6.3×10^2	1.1×10^7	0.44×10^{15}	1500
Glass ball	Hirono, Uji, Kyoto Pref.	6.3	4.8×10^5	2.31×10^{15}	1500
Zircon (baked earth)	Iwasaki, Nisshin, Aichi Pref.	2.3×10^3	4.7×10^7	0.44×10^{15}	1300
"Tatara"	Ichinomiya, Okayama Pref.	19	2.6×10^6	2.81×10^{15}	1300
Zircon (baked earth)	Sakai, Osaka Pref.	2.7×10^3	6.3×10^7	0.46×10^{15}	1200
Zircon (tile)	Nagaoka, Kyoto Pref.	5.2×10^2	1.3×10^7	0.50×10^{15}	1200
		5.2×10^2	1.3×10^7	0.50×10^{15}	1200
		4.9×10^2	1.2×10^7	0.50×10^{15}	1150
Zircon (pottery)	Nagaoka, Kyotō Pref.	4.3×10^2	1.1×10^7	0.50×10^{15}	1150
"Tatara"	Habutenno, Wake, Okayama Pref.	14	1.1×10^6	1.47×10^{15}	1150
Zircon (opening of folge)	Arai, Fukaya, Siraishi City	4.3×10^2	1.1×10^7	0.50×10^{15}	1150
"Tatara"	Michikunihara, Fukaya, Shiraishi City	14	2.1×10^6	2.81×10^{15}	1150
		13	2.1×10^6	2.80×10^{15}	1050
		13	2.1×10^6	2.82×10^{15}	1050
"Tatara"	Habutenno, Wake, Okayama Pref.	9.3	1.5×10^6	2.81×10^{15}	1050
		10	1.5×10^6	2.80×10^{15}	1150
		9.4	1.5×10^6	2.82×10^{15}	1100
"Tatara"	Odatemori, Ajigazawa, Aomori Pref.	7.8	9.8×10^5	2.82×10^{15}	700
Zircon (baked earth)	Fuso, Aichi Pref.	1.4×10^3	5.5×10^7	0.44×10^{15}	700
Glaze on bowl	Isumi, Toki City	0.45	1.1×10^5	1.9×10^{15}	400

Publication Delay

SIR,—Delay in the publication of manuscripts in specialized scientific journals is becoming more and more frustrating because of the pace of science today. In many domestic journals it takes 6–9 months to publish a manuscript after receipt. In the case of European journals, it takes at least 2 more months because of postal handling of manuscripts and galley proofs; there can be even further delay if the manuscript has to be revised. In addition to this, journals can take about 2 months by surface mail to travel between Europe and the United States.

This means that 8–11 months may elapse before a submitted manuscript reaches the scientific community. This delay is frustrating for two reasons: (i) the work may be needlessly duplicated by the efforts of colleagues which might more profitably have been spent on the other aspects of the same problem; and (ii) the scientific and social impact of the information may be diminished.

Several solutions to this problem are available. (i) Journals could publish more manuscripts in each issue, change to a larger page size, or publish more frequent issues. Some periodicals have already adopted one or more of these solutions. (ii) Reviewing procedures

could be made more efficient; more European journals could let their US editors review North American manuscripts and vice versa; (iii) mailings could be made by air; (iv) journals (which do not already do so) could make available a list of accepted manuscripts along with the authors' names, addresses, and expected dates of publication. This list could either be published by the journal itself or by some scientific abstracting or indexing service (for example, *Biological Abstracts*, *Chemical Abstracts*, *Current Contents*) and the pre-publication manuscripts could be obtained either directly from the authors or, at a reasonable cost, from the particular journal.

We are aware that each of these suggestions entails additional publication costs. However, we believe that the fourth suggestion should be considered because it could greatly diminish the communication delay with the minimal additional publication expense.

Yours faithfully,

M. GASSMAN
D. PENNEY
M. A. Q. KHAN

*Department of Biological Sciences,
University of Illinois at Chicago Circle,
College of Liberal Arts and Sciences*

How to Succeed . . . ?

We have received the following chain letter from an offended reader.

Dear Doctor,

Publish or perish? Today, more scientists than ever before are faced with this problem. This letter brings you the opportunity not only to publish but to appear as a co-author of many interesting and important articles bearing on research problems in a variety of scientific disciplines. Naturally, we want to help you, but in order to do this you have to help us.

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Send the eight copies to eight well-known scientists of your acquaintance. Try to maintain as far as possible the high standard of scientific excellence represented on the present list.

If you send eight copies off within a few weeks and publish, as you usually do, within a few months, you should by the end of a year be the co-author of approximately 16,000,000 first-class papers. Not bad going—Eh!

Obituary

Dr H. Barrell

HARRY BARRELL, who died on February 16, 1972, was a former Superintendent of what was then the Standards Division of the National Physical Laboratory. His reputation was international and he was for many years one of the leading metrologists who brought about the replacement of mechanical standards of length and time by the present atomic standards, the wavelength of a line in the spectrum of krypton-86 and the caesium-133 atomic frequency standard.

Barrell, whose family came from Herefordshire, took his BSc degree at Imperial College and then worked for a time with R. H. Fowler, the spectroscopist. He was appointed to the staff of the Metrology Division in 1923 and between the wars worked with J. E. Sears on the measurement of the metre in terms of the wavelength of the cadmium red line, and on the refractive index and dispersion of air; the latter work is definitive.

The distance over which interference fringes can be obtained with the red line in the spectrum of cadmium is

much less than 1 m and therefore, much as in the work of Fabry and Perot and others, Sears and Barrell used a system of fringes of interference in white light to relate a length of one ninth of a metre, directly measurable by interference in the cadmium red line, to a length of 1 m in two stages of comparison in white light. The experiments were characterized by imaginative design and careful execution, supported by the excellent facilities of the workshops of the National Physical Laboratory.

In the primary work, the length of a mechanical end standard about 1 m long was determined in a vacuum by an interferometer in terms of the cadmium line; at the time the result seemed discordant with that of parallel determinations in Germany but later studies of the properties of the light sources showed that the agreement was in fact very close. It was, however, clear that the discrepancies between these and other determinations of the metre arose mainly from the errors in making microscope settings upon the line standards of length by which the metre was then defined. Accordingly, when after 1945 single isotopes became avail-

able, it was decided that there was no point in making further comparisons with the mechanical standards but that, instead, the mean value of the metre in terms of the cadmium line should be accepted and should be converted to a value in terms of a single isotope line through purely interferometric comparisons of the wavelength of the latter with that of the cadmium line. Barrell was one of the first to study the radiation from single isotopes, making use of the refined interferometric techniques he had developed and, in particular, he made a careful study of the dependence of the wavelengths of lines in the spectrum of mercury-198 upon the conditions of excitation.

He was prominent in the international discussions that led to the adoption of the krypton-86 line and succeeded J. E. Sears as a member of the International Committee for Weights and Measures. Meanwhile Dr L. Essen was developing the caesium atomic beam standard of frequency, and Barrell, as a member of the International Committee and as President of its Advisory Committee on the Definition of the Second, had a decisive effect on the adoption of the present

definition of the second in terms of a hyperfine transition in caesium-133.

Barrell's work on atomic standards has been a major contribution to the transformation that methods and standards of measurement have undergone in the past 30 years, leading to much improved precision and accessibility of standards, with all that implies for reliability, convenience and economy in physics and engineering.

He himself made notable contributions to engineering applications, especially of interferometric methods for calibrating engineers' gauges.

As Superintendent of the Standards Division, Barrell brought together into an effective team many groups working on problems of measurement in the National Physical Laboratory and he particularly encouraged the first steps that were taken to relate electrical standards to measurement of frequency. He was an inspiring leader of his division and many important pieces of work depended greatly on his encouragement. The high regard in which the metrological work of the National Physical Laboratory is held internationally is the result of the foundations Barrell and his predecessors laid in their choice of programmes and people.

In his younger days Barrell was a keen member of the National Physical Laboratory hockey club, the standing of

which has always been high; in later years he took up amateur acting and shone in character parts, for example in "The Crucible" and "The School for Scandal".

Distinguished scientist, international leader, inspiring Superintendent as he was, by most of those who knew him Barrell will be remembered for his encouragement and support but above all for his friendship.

Announcements

University News

Professor S. J. Prias, National Institute of Economic and Social Research, has been appointed visiting professor in econometrics in the Department of Social Science and Humanities, **The City University**.

Professor Phillip A. Griffiths has been appointed professor of mathematics in **Harvard University**. **Dr. George Brooks Field** has been appointed professor of astronomy at Harvard, and will succeed **Professor Alexander Dalgarno** as director of the Harvard College Observatory in 1973.

Dr A. C. T. North has been appointed professor of molecular biophysics in the

Astbury Department of Biophysics, **University of Leeds**.

Professor E. A. Bell, University of Texas, has been appointed to the chair of biology tenable at King's College, **University of London**. The title of professor of zoology has been conferred on **Dr J. L. Cloudsley-Thompson**, in respect of his post at Birkbeck College, and that of professor of haematology on **Dr J. G. Humble**, in respect of his post at the Westminster Medical School.

Professor Colin Kaplan, head of the Microbiology Department, **University of Reading**, has been appointed dean of the Faculty of Science.

Dr J. D. Woods, Meteorological Office, Bracknell, has been appointed to the chair of physical oceanography at the **University of Southampton**.

Appointments

Professor J. Wreford Watson, University of Edinburgh, has been appointed a member of the **Social Science Research Council**.

Dr Eric J. Gibson has been appointed an assistant director of the Building Research Establishment and head of **Princes Risborough Laboratory**.

Continued from p. 19

therapy for reasons which are not clear at present. This group includes many patients with brain tumours, soft tissue sarcoma, adenocarcinoma of the stomach and intestine and some melanomas. Locally advanced carcinoma of the cervix and bladder are also rarely cured by existing methods, and the recurrence of these tumours after treatment presents an extremely difficult problem which is not amenable to further radiotherapy or chemotherapy. If the cause of failure lies in the unfavourable radiobiological properties of radiation with low LET, use of radiation of higher LET might make a significant difference and certain groups might show a dramatic response to the new form of treatment. It is clear, however, that the possible advantages of radiation with high LET are of a speculative kind, and in any individual case they may instead turn out to be disadvantages. The presence of hypoxic cells in many tumours is the only reason for which radiation with high LET is certain to be advantageous, and even this may be an illusion because fractionation of low LET therapy may of itself eliminate the resistant hypoxic component¹⁶. It is to be hoped that the trial of fast neutron therapy at present in progress at the Hammersmith Hospital¹⁷ will in due course answer the question of whether radiation with high LET has any radiotherapeutic advantage.

It is important to notice, however, that the case for heavy ions and mesons does not rest solely on the high LET of the radiation in the region of the peak. The distribution with depth of biologically effective dose is so very much closer to the ideal than anything which can be achieved with X-rays or electrons that improved therapeutic results must follow from this alone. This will be most marked in the case of very deep-seated cancers such as those of the bladder and the uterus.

It has by now become clear to everyone in the field of

cancer research that the idea of a single wonder drug—a "cure for cancer"—is a mirage. Progress in the control of this group of diseases has taken the form of many small steps, each giving improvement in one facet of the problem and all taken together representing an important advance. Introduction of radiotherapy with heavy ions or negative pions will be a substantial step on the road.

I thank many of my colleagues for advice about several aspects of this article.

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Dr W. Stanley Brown, Bell Laboratories, Arthur W. Elias, 3i Company/Information Interscience Incorporated, **F. G. Kilgour**, Ohio College Library Center, **Mrs M. S. Morris**, Allied Chemical Corporation, and **Dr Harrison Shull**, Indiana University, have been appointed to the **Chemical Abstracts Service Advisory Board**, and **John Griffith**, IBM Corporation, has been reappointed.

Miscellaneous

Alfred P. Sloan Foundation research fellowships have been awarded to the following young scientists: **B. R. Barrett**, University of Arizona (physics); **J. E. Gunn**, California Institute of Technology (astrophysics); **Hans C. Andersen**, Stanford University (chemistry); **C. Denson Hill**, Stanford University (mathematics); **A. M. Nur**, Stanford University (geophysics); **V. Petrosian**, Stanford University (astrophysics); **A. Kas**, University of California, Berkeley (mathematics); **T. Lam**, University of California, Berkeley (mathematics); **R. McKenzie**, University of California, Berkeley (mathematics); **H. F. Schaefer**, University of California, Berkeley (chemistry); **J. I. Silk**, University of California, Berkeley (astrophysics); **G. N. LaMar**, University of California, Davis (chemistry); **V. L. Trimble**, University of California, Irvine (astrophysics); **D. A. Evans**, University of California, Los Angeles (chemistry); **S. J. Putterman**, University of California, Los Angeles (physics); **D. S. Sigman**, University of California, Los Angeles (biochemistry); **R. L. Vold**, University of California, San Diego (chemistry); **J. C. Wheeler**, University of California, San Diego (chemistry); **M. Shub**, University of California, Santa Cruz (mathematics); **L. L. Miller**, Colorado State University (chemistry); **W. C. Lineberger**, University of Colorado (chemistry); **J. E. Faller**, Wesleyan University, Middletown (astrophysics); **L. Corwin**, Yale University (mathe-

matics); **R. P. Geroch**, University of Chicago (mathematical physics); **D. Chandler**, University of Illinois (chemistry); **S. Chang**, University of Illinois (physics); **R. P. Kaufman**, University of Illinois (mathematics); **W. O. McClure**, University of Illinois (neurochemistry); **J. T. Yardley**, University of Illinois (chemistry); **A. Weitsman**, Purdue University (mathematics); **D. N. Coucouvanis**, University of Iowa (chemistry); **W. C. Stwalley**, University of Iowa (chemistry); **D. C. Polahd**, Johns Hopkins University (chemistry); **S. Kerpel-Fronius**, University of Maryland (neurochemistry); **B. M. Zuckerman**, University of Maryland (radioastronomy); **R. C. Clarke**, Boston University (chemistry); **R. A. Laursen**, Boston University (bioorganic chemistry); **D. I. Lieberman**, Brandeis University (mathematics); **M. Schachner**, Harvard Medical School (neurobiology); **G. R. Kempf**, Harvard University (mathematics); **J. A. Osborn**, Harvard University (chemistry); **J. E. Pilcher**, Harvard University (physics); **W. P. Reinhardt**, Harvard University (chemistry); **C. M. Bender**, MIT (applied mathematics and theoretical physics); **J. W. Negele**, MIT (physics); **D. A. Garelick**, Northeastern University (physics); **R. B. Hallock**, University of Massachusetts (physics); **W. C. McHarris**, Michigan State University (nuclear chemistry); **A. J. Ashe**, University of Michigan (chemistry); **B. J. Evans**, University of Michigan (crystallography); **M. Raban**, Wayne State University (chemistry); **F. P. Calaprice**, Princeton University (physics); **W. Carithers**, Columbia University (physics); **J. L. Gross**, Columbia University (mathematics); **R. W. Guernsey**, Columbia University (physics); **A. J. Chorin**, New York University (applied mathematics); **R. E. Zigmond**, Rockefeller University (neurochemistry); **D. M. Hanson**, State University of New York (chemistry); **S. J. Osher**, State University of New York (mathematics); **F. H. Shu**, State

University of New York (astrophysics); **S. R. Simon**, State University of New York (biochemistry); **G. D. Sprouse**, State University of New York (physics); **J. W. Kalat**, Duke University (neuropsychology); **P. J. Kropp**, University of North Carolina (chemistry); **J. F. W. Keana**, University of Oregon (chemistry); **T. W. Mihalisin**, Temple University (physics); **R. Eisenberg**, Brown University (chemistry); **S. Ying**, Brown University (physics); **F. R. Harvey**, Rice University (mathematics); **D. J. Kouri**, University of Houston (chemistry); **C. N. Davids**, University of Texas (nuclear astrophysics); **E. G. Adelberger**, University of Washington (physics); **N. H. Andersen**, University of Washington (chemistry); **E. Fetz**, University of Washington (neurophysiology); **M. H. O'Leary**, University of Wisconsin (chemistry); **A. O. W. Stretton**, University of Wisconsin (molecular neurobiology); **B. C. Eu**, McGill University (chemistry); **I. Ozier**, University of British Columbia (physics); **D. R. Arnold**, University of Western Ontario (chemistry).

George Waterston, regional officer for Scotland of the **Royal Society for the Protection of Birds**, has been awarded the Society's gold medal.

Professor A. J. P. Martin, Wellcome Foundation Limited, has been awarded **Japan's Order of the Rising Sun**.

Erratum

IN the article entitled "Mechanism of Immunologically Specific Killing of Tumour Cells by Macrophages" by R. Evans and P. Alexander (*Nature*, **236**, 168; 1972), the reference number in line 11 should be to reference 4 in the bibliography; similarly in line 12 from the bottom of page 170, the reference number should be to reference 9 in the bibliography. Reference 8 in the bibliography should read: *Clin. Exp. Immunol.*, **6**, 1 (1970).

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